A

Abbreviations
Summary
Samenvatting
Curriculum Vitae
Ph.D. Portfolio
Publications
Acknowledgements

ABBREVIATION INDEX

A-OBs	A-OBs: Activin A-treated osteoblasts
activin-ECM	Activin-treated extracellular matrix
ACVR2A	Activin receptor type 2A
ACVR2B	Activin receptor type 2B
ACVR1B/ALK4	Activin receptor type 1B
ADD1	Adducin-1
AFM	Atomic force microscopy
AHSG	Alpha-2-HS-glycoprotein
AKT/PKB	AKT serine threonine kinase (protein kinase B)
ALP(L)/TNAP	Alkaline phosphatase, tissue-nonspecific
alpha-MEM	Alpha minimum essential medium
ANGPTL4	Angiopoietin like 4
ANO6	Anoctamin6
ANOVA	Analysis of variance
ANXA1	Annexin A1
ANXA11	Annexin A11
ANXA2	Annexin A2
ANXA5	Annexin A5
ANXA6	Annexin A6
ANXA7	Annexin A7
AP-1/JUNB	Activator protein 1/JunB proto-oncogene
APC	Axin/adenomatous polyposis coli
ARIPs	Activin receptors interacting proteins
ARS	Alizarin red staining
ATF4	Activating TF4
AUC	Area under curve
β-TCP	Beta-tricalcium phosphate
BGLAP	Osteocalcin
BMP2	Bone morphogenic protein 2
BMP2K	BMP2 inducible kinase
BMP4	Bone morphogenic protein 4
BMPR-I	Bone morphogenic protein receptor type 1
BMPR-II	Bone morphogenic protein receptor type 2
BMPs	Bone morphogenic proteins
BTE	Bone tissue engineering
BSP	Bone sialoprotein
CAT	Catalase
cGMP	Current good manufacturing practices
COL12A1	Collagen type XII alpha 1

COL1A1	Collagen type I alpha 1
COL3A1	Collagen type III alpha 1
COMP	Cartilage oligomeric matrix protein
COS7	Fibroblast cell line, monkey-derived
CST3	Cystatin C
CTGF	Connective tissue growth factor
CTHRC1	Collagen triple helix repeat containing 1
CTSZ	Cathepsin Z
CYR61	Cysteine-rich protein 61
DAPI	4,6-diamino-2-phenylindole
DAVID	Database for Annotation, Visualization and Integrated Discovery
DLX5	Distal-less homeobox-5
DMP1	Dentin matrix protein
DMSO	Dimethyl sulfoxide
DNase	Deoxyribonuclease
ECM	Extracellular matrix
EDN1	Endothelin 1
EGF	Epidermal growth factor
EGFR	Epidermal growth factor receptor
ENPP1	Ectonucleotide pyrophosphatase/phosphodiesterase family member 1
EPO	Erythropoietin
ER	Endoplasmic reticulum
ERK	Extracellular signal-regulated kinase
FACS	Fluorescence-activated cell sorting
FAK	Focal adhesion kinase
FBLN1	Fibulin-1
FBLN2	Fibulin-2
FBLN5	Fibulin-5
FBN1	Fibrillin 1
FBS	Fetal bovine serum
FETUB	Fetuin B
FGF23	Fibroblast growth factor 23
FGFR3	Fibroblast growth factor receptor 3
FITC	Fluorescein isothiocyanate
FN	Fibronectin
FST	Follistatin
FZD	Frizzled receptor
GAPDH	Glyceraldehyde 3-phosphate dehydrogenase
GFs	Growth factors
GO	Gene Ontology
GSK-3β	Glycogen synthase kinase 3 beta

GTP	Guanosine triphosphate
HA	Hydroxyapatite
HA-TCP	Hydroxyapatite-tricalcium phosphate beta
HAPLN1	Hyaluronan proteoglycan link protein 1
HDAC	Histone deacetylase
hESCs	Human embryonic stem cells
HIF	Hypoxia inducible factor
HSCs	Hematopoietic stem cells
HSPA5	Heat shock protein family A member 5
HSPG2	Heparan sulfate proteoglycan core protein
hTERT	Telomerase reverse transcriptase
ID1	Inhibitor of DNA binding 1
IDs	Identifiers
IGF	Insulin-like growth factor
IGFBP3	Insulin-like growth factor-binding protein 3
IGFBP5	Insulin-like growth factor-binding protein 5
IGFBP7	Insulin-like growth factor-binding protein 7
IHH	Indian hedgehog
ILK	Integrin linked kinase
IPA	Ingenuity pathway analysis
IR	Infrared
JNK	c-Jun N-terminal kinase
kDa	Kilodalton
KLF10	Krupper-like factor 10
LAMC1	Laminin subunit gamma-1
LAP	Latency associated peptide
LC-MS	Liquid chromatography–mass spectrometry
LEF	Lymphoid enhancer binding factor
LFQ	Label-free quantification
LRP5/6	Lipoprotein receptor-related protein 5 and 6
LTPBs	Latent TGF beta binding proteins
LY	LY294002
M-CSF	Macrophage colony-stimulating factor
M-PER	Mammalian protein extraction buffer
MAPK	Mitogen-activated protein kinase
MAPK1	Mitogen-activated protein kinase 1
MEPE	Matrix extracellular phosphoglycoprotein
MGP	Matrix gla protein
min-ECM	Mineralizing extracellular matrix
miRNA	MicroRNA
MMA	Methyl methacrylate

MMP14	Matrix metalloproteinase 14
MMPs	Matrix metalloproteinases
mRNA	Messenger RNA
MS	Mass spectrometry
MSCs	Mesenchymal stem/stromal cells
MSX-2	Msh-homeobox homologue-2
MT1-MMP	Membrane type 1 matrix metalloproteinase
NCPs	Non-collagenous proteins
NF-kB	Nuclear factor kappa-light-chain-enhancer of activated B cells
nonmin-ECM	Non-mineralizing extracellular matrix
NOX4	NADPH oxidase 4
NSG	Non-obese diabetic/Scid IL2R gamma
NT-OBs	Not treated osteoblasts
OBs	Osteoblasts
OPG	Osteoprotegerin
OPN	Osteopontin
OSX	Osterix
p38	p38 mitogen-activated protein kinase
PAK	p21-activated kinase
PARP	Poly ADP-ribose polymerase
PBS	Phosphate-buffered saline
PDGF	Platelet-derived growth factor
PFA	Paraformaldehyde
PHEX	Phosphate-regulating gene with homologies to endopeptidases on the X-chromosome
PI	Propidium iodide
PI3K	1-phosphatidylinositol 3-kinase
PI3KR1	Phosphatidylinositol 3-kinase regulatory subunit alpha
PKA	Protein kinase A
PKC	Protein kinase C
PLA	Poly-lactic acid
PLL	Poly-L-Lysine
POSTN	Periostin
PPARG	Peroxisome proliferator activated receptor gamma
PTC	Patched
PTEN	Phosphatase and tensin homolog
PTH	Parathyroid hormone
PTPN1	Tyrosine-protein phosphatase non-receptor type 1
qPCR	Quantitative polymerase chain reaction
Ra	Roughness Average
RAP1A	Ras-related protein Rap1a

RANKL	RANK ligand
RGD	Arg-Gly-Asp
RNA	Ribonucleic acid
ROCK	Rho-associated protein kinase
RUNX	Runt-related transcription factor 2
SD	Standard deviation
SDS-PAGE	Sodium dodecyl sulphate polyacrylamide gel electrophoresis
SEM	Scanning electron microscopy
SIBLING	Small integrin binding ligand N-linked glycoprotein
SMAD1/5/8	SMAD family member 1/5/8
SMAD2/3	SMAD family member 2/3
SMAD4	SMAD family member 4
SMAD7	SMAD family member 7
SMO	Smoothened
SOD1	Superoxide dismutase 1
SOD2	Superoxide dismutase 2
SOST	Sclerostin
SPARC/SPOCK1	Osteonectin
SPF	Pathogen free facility
SSCs	Skeletal stem cells
SV-HFOs	Simian virus 40-immortalized human osteoblast cell line
TβR-I	Transforming growth factor beta receptor type 1
TβR-II	Transforming growth factor beta receptor type 2
TBS	Tris-buffered saline
TCF	T cell factor
TGF-β	Transforming growth factor beta
TGFB1	Transforming growth factor beta 1
TNC	Tenascin C
TNF-α	Tumor necrosis factor alpha
TSP1/THBS1	Thrombospondin 1
TWIST1	Twist homolog 1
U	Enzyme unit
UPR	Unfolded protein response
VCAN	Versican
VEGF	Vascular endothelial growth factor
VIM	Vimentin
Vmax	Maximal velocity
VTN	Vitronectin
WM	Wortmannin
Wnt	Wingless type-MMTV integration site family

SUMMARY

Bone is a very dynamic tissue composed of bone extracellular matrix (ECM) in a tight interplay with bone cells, such as osteoblasts, the bone-forming cells that secrete and mineralize bone ECM. Extracellular signaling molecules and the ECM with its biochemical and biophysical cues continuously influence bone cell behaviour, tightly regulating bone formation and bone quality. Bone tissue is continuously undergoing remodelling and can self-repair after fracture healing. However, major challenges remain in case of large skeletal defects such as large fractures, bone metastasis, or age-related bone fragility. Bone tissue engineering has recently emerged as therapeutic strategy in alternative to bone grafts. Understanding how bone ECM and signaling molecules influence cell behaviour is crucial to ameliorate mesenchymal stromal cell (MSC) properties and to develop optimal bone grafts that reproduce the physiological bone microenvironment for bone tissue engineering applications. Cell-derived ECMs have been proposed as a good alternative to native decellularized organs in the context of tissue engineering, in combination with scaffolds, to reproduce the physiological architectural complexity of ECM. In this thesis, we created three osteoblast-derived ECMs and we investigated how they affect MSC osteogenic differentiation and mineralization, to ameliorate the promising properties of MSCs for bone regeneration. Moreover, we studied how the extracellular environment with signaling molecules such as Activin-A affects ECM composition and mineralization, thereby modulating bone tissue quality.

In **Chapter 2**, we presented an osteoblast-derived ECM, that was able to enhance the adhesion of the MSCs, and increased the percentage of proliferating cells more than 2-fold. The ECM was osteopromotive for MSCs both *in vitro* and *in vivo*, in a mouse model of ectopic bone formation. More than 50% of the proteins were shared with the ones of human bone samples, revealing a high homology with the *in vivo*-like microenvironment. Known ECM structural proteins such as COL1A1, FN1, TNC were detected among the most abundant proteins by mass spectrometry, together with known mediators of mineralization such as annexins, revealing the essential role of ECM in mediating the effect we observed. In addition to core matrisome components, we detected also antioxidant and cytoskeletal proteins, that need further investigation in the context of matrix mineralization.

In **Chapter 3**, we modified the composition of the osteoblast-derived ECM presented in Chapter 2 (min-ECM), and obtained ECMs with different mineralization phenotypes. The nonmin-ECM was obtained by culturing MSCs without osteogenic inducers, and had a reduced osteogenic potential than the min-ECM. The comparative proteomic analysis confirmed that proteins involved in mineralization such as Annexins and Alkaline phosphatase were downregulated in the nonmin-ECM,

whereas structural and ribosomal proteins were upregulated. We highlighted the use of this ECM not only to study candidates involved in mineralization, but also to modulate MSC behaviour to regulate bone quality.

Signaling molecules also modulate osteoblast behaviour in bone. In this context, Activin-A was previously shown to inhibit matrix mineralization, though the mechanism remains largely unknown. To get further insights in Activin A-mediated inhibition of mineralization, in **Chapter 3** we showed that an Activin-ECM obtained by culturing MSCs with osteogenic inducers and Activin-A, was still able to retain Activin A-activity in reducing matrix mineralization. Comparative proteomic analysis with the min-ECM presented in Chapter 2 showed that Activin was able to modify the ECM composition, by upregulating extracellular proteins and downregulating mitochondrial and membrane proteins. Moreover, in **Chapter 4** we showed that 2-day treatment with Activin-A was sufficient to inhibit matrix mineralization in human osteoblasts. Microarray data revealed that osteoblast gene expression was altered in a 2-wave fashion over time. Genes involved in the first wave of gene expression changes (within 8 hours after Activin A-start) were mainly transcription factors, whereas ECM proteins were the target of second wave changes (1-2 days after Activin A-start). Moreover, we showed that Activin-A altered the miRNA profile of osteoblasts, including miRNAs that target genes involved in TGF-β signaling. Overall, our findings showed that Activin-A alters matrix composition, resulting in a matrix that fails to support the later stages of osteoblast differentiation, confirming our previous work that illustrated that Activin-A strongly inhibited matrix mineralization. We showed that Activin-A modulates osteoblast differentiation, by influencing osteoblast miRNA profile, gene transcription and eventually altering matrix mineralization, giving further insights in unravelling Activin-A mechanism of action and promoting the use of Activin-A to modulate osteoblast behaviour to regulate bone quality.

The physical cues of the ECM are mechanosensed by bone cells via integrin-mediated signaling. This activates an intricate network of kinase signaling cascades, which lies at the core of signal transduction that regulates cellular behaviour. However, the relay of information from integrin-engagement to modulation of cell physiology is still not fully known. In **Chapter 5** we employed a kinase array to disentangle MSC adhesion to the osteoblast-derived ECM presented in Chapter 2 in comparison to titanium. Titanium surface has good mechanical properties and can be tailored in porosity to increase surface area and cell adhesion, representing an optimal scaffold already used for bone tissue engineering applications. We showed that MSCs on both surfaces exhibited substantial overlapping activated kinase signatures, but with a higher level of kinase activity in MSCs on ECM. Moreover, we successfully used the peptide array to select PI3K/Akt signaling and confirmed its importance

in MSC viability and adhesion. Without *a priori* assumptions, kinase arrays helped in the study of cell/surface interplay. Our osteoblast-derived ECM could be used as coating to ameliorate the properties of titanium, and for the rational design of novel and cell-instructive scaffolds for tissue engineering.

In conclusion, the findings described in this thesis give insights into how the extracellular microenvironment influences cell behaviour and how we can manipulate it to ameliorate stromal cell properties for regenerative purposes. We took advantage of biochemical analyses combined with various -omics data to characterize the devitalized ECMs that we produced and to study the effect on osteoblast behaviour, with a focus on mineralization. In **Chapter 6** we presented the limitations of this system together with the potential applications to ameliorate *in vitro* cultures, to improve the promising properties of mesenchymal stromal cells in regenerative medicine.

SAMENVATTING

Bot is een erg dynamisch weefsel dat voor een groot gedeelte bestaat uit extracellulaire matrix (ECM) eiwitten en calciumfosfaat. Deze ECM eiwitten worden geproduceerd door osteoblasten, de botvormende cellen. De verkalking van de ECM zorgt vervolgens voor de stevigheid van het bot en skelet. Botweefsel is continu aan het veranderen door een samenspel van botvormende en -afbrekende cellen. Dit process is essentieel voor de reparatie en genezing van fracturen. Extracellulaire signaalmoleculen en de biochemische en biofysische eigenschappen van de ECM kunnen het gedrag van deze bot cellen beïnvloeden en zo ook de vorming, kwaliteit en stevigheid van het bot bepalen. Tot op heden blijft het een grote uitdaging om gebreken zoals grote breuken en aandoeningen, veroorzaakt door botmetastases en leeftijdsgebonden afname van de botmassa, te genezen. Tissue engineering zou een goed alternatief kunnen zijn voor het gebruik in bot regeneratieve geneeskunde en de genezing van deze aandoeningen aan het bot.

Voor het ontwikkelen van bot tissue engineering is het van belang deze cellen te bestuderen in een vergelijkbare bot omgeving en zo de invloed van de ECM en signaalmoleculen op het gedrag van botvormende cellen te bepalen. *In vitro* geproduceerd ECM zou gebruikt kunnen worden in bot tissue engineering om zo met een dragermateriaal de complexe fysiologische samenstelling en architectuur van het bot te reproduceren. Om het gebruik van mesenchymale stromale cellen (MSCs) in regeneratieve geneeskunde en tissue engineering te verbeteren hebben we in dit proefschrift 3 soorten ECMs gemaakt, geproduceerd door osteoblasten, en hebben onderzocht hoe deze ECMs de osteoblast differentiatie en mineralisatie van MSCs kunnen reguleren. Ook hebben we onderzocht hoe deze extracellulaire omgeving en samenstelling wordt beïnvloed als de osteoblasten die deze ECM maken worden behandeld met Activin-A.

In **hoofdstuk 2** onderzochten wij de devitalisatie van een ECM die door osteoblast gedifferentieerde MSCs (min-ECM) is geproduceerd. Deze min-ECM was in staat om de hechting van MSCs aan deze ECM te versnellen. Verder vonden wij 2 keer zoveel prolifererende cellen wanneer we deze lieten groeien op een gedevitaliseerde min-ECM en stimuleerde deze de osteogene differentiatie *in vitro* én in een muismodel voor ectopische botvorming. Analyses van het proteome lieten zien dat de samenstelling van de *in vitro* gegenereerde ECM vergelijkbaar was met de ECM die voorkomt in het bot. Meer dan 50% van de eiwitten in de ECM waren ook terug te vinden in humane bot biopsies. Tussen de meest abundante eiwitten vonden we bekende structurele eiwitten van het ECM, zoals Collageen 1 (COL1A1), Fibronectine (FN1) en Tenascin C (TNC), maar ook annexines die een belangrijke rol spelen in de mineralisatie van de ECM. Naast de vele bekende ECM componenten, konden we

ook antioxidanten en cytoskelet eiwitten identificeren. Verder onderzoek is nodig om te bepalen wat de functie is van deze eiwitten is in de mineralisatie van de ECM.

In **hoofdstuk 3**, hebben we vervolgens onderzocht wat er gebeurd met de osteoblast differentiatie zodra we de samenstelling van de ECM uit hoofdstuk 2 veranderde. Hiervoor hebben we 2 verschillende ECMs gebruikt: 1) nonmin-ECM en 2) act-ECM. De nonmin-ECM werd verkregen door het groeien van ongedifferentieerde MSC's en de act-ECM werd verkregen door het groeien van osteoblast differentiërende MSC's die behandeld waren met Activin-A. Beide ECMs hebben een ander mineralisatie fenotype in vergelijking met de min-ECM die beschreven is in hoofdstuk 2. De nonmin-ECM remde de osteoblast differentiatie van MSCs en proteomics analyse bevestigde vervolgens dat de eiwitten die betrokken zijn bij de mineralisatie, zoals annexines en alkalische fosfatase, minder verrijkt waren in de nonmin-ECM, terwijl structurele en ribosomale eiwitten in grotere hoeveelheden aanwezig waren. Hoewel het mechanisme nog onbekend is, is in eerder onderzoek aangetoond dat Activin-A de mineralisatie van osteoblast differentierende MSCs kan remmen. Wanneer osteoblast differentiërende MSCs werden gegroeid op de act-ECM zagen we dat de matrix mineralisatie verminderd was ten opzichte van min-ECM. Vergelijking van het proteome liet zien dat Activin-A de ECM kan moduleren door de inductie van extracellulaire eiwitten en de remming van mitochondriale- en membraan-eiwitten.

Vervolgens hebben we in **hoofdstuk 4** onderzocht wat het effect op de mRNA en miRNA expressie is zodra we osteoblast differentiërende MSCs behandelen met Activin-A. Een 2-daagse Activin-A behandeling van MSCs was voldoende om de mineralisatie van osteoblasten te remmen. De microarray experimenten lieten zien dat de genexpressie veranderingen in 2 verschillende fases plaatsvonden. In de eerste fase, de eerste 8 uur na toevoeging van Activin-A, veranderde voornamelijk de expressie van verschillende transcriptiefactoren. Terwijl in de tweede fase, 1-2 dagen na Activin-A inductie, genen veranderde die coderen voor ECM eiwitten. Bovendien vonden wij dat de expressie van verschillende miRNAs door Activin-A werden gereguleerd, zoals de expressie van miRNAs die betrokken zijn in de regulatie van genen in de TGF-β signaalcascade. De genoomwijde genexpressie analyse laten zien dat Activin-A de mRNA en miRNA expressie in osteoblast differentieren MSCs kan veranderen en daaropvolgend een veranderde samenstelling van de ECM en remming van de mineralisatie veroorzaakt.

De signalen van de ECM worden door botcellen via integrin-gemedieerde signalen doorgegeven. Deze signalering activeert een complex netwerk van kinases en signaaltransductie cascades, die samen het gedrag van cellen kunnen reguleren. Echter, het is nog niet helemaal duidelijk welke cascades worden geactiveerd zodra MSC hechten aan een ECM en hoe deze de osteoblast differentiatie kan reguleren.

In **hoofdstuk 5** hebben we gebruik gemaakt van peptide arrays om te bepalen welke signaaltransductie cascades worden geactiveerd zodra MSC hechten aan een ECM geproduceerd door osteoblasten en hebben deze vergeleken met cellen die hechten aan een titanium oppervlakte. Titanium is een dragermateriaal dat al veel gebruikt wordt in bot tissue engineering toepassingen en regeneratieve geneeskunde. Het heeft vergelijkbare mechanische eigenschappen als bot en kan eenvoudig worden aangepast in de porositeit van het oppervlak om hechting van cellen te verhogen. We laten zien dat beide oppervlakte, min-ECM en titanium, voor een groot gedeelte dezelfde kinases activeren en deactiveren maar op min-ECM met een hogere activiteit. Bovendien hebben we de PI3K / Akt signalering gevonden en bevestigen het belang van deze cascade in de hechting en viabiliteit van MSCs aan dit oppervlak. Deze kinase arrays hebben geholpen om signalerings cascades te identificeren die worden gereguleerd zodra MSCs hechten aan een oppervlakte. Verder laten we door middel van deze experimenten zien dat de min-ECM gebruikt kan worden als coating om de eigenschappen van titanium te verbeteren en voor verder gebruik en ontwerp van nieuw drager materiaal in bot tissue engineering.

Samenvattend, de in dit proefschrift beschreven bevindingen zullen meer kennis geven over hoe de extracellulaire omgeving, zoals de extracellulaire matrix, het gedrag van osteoblasten kunnen beïnvloeden en hoe we deze kunnen manipuleren om het gebruik van MSCs in de regeneratieve geneeskunde te verbeteren. In **hoofdstuk 6** bediscussiëren wij de beperkingen van het onderzochte systeem alsmede de mogelijke toepassingen van de ECM in bot tissue engineering en het gebruik van MSCs in regeneratieve geneeskunde.

CURRICULUM VITAE

Marta Baroncelli

17-01-1987 Born in Prato, Italy

2006-2009 B.Sc. in Biotechnology
University of Florence, Italy

2009-2011 M.Sc. in Medical and Pharmaceutical Biotechnology
University of Florence, Italy
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PHD TRAINING ACTIVITIES

Courses and Workshops

2012 Biomedical research technique course
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2013 Molecular medicine course
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Basic and translational endocrinology course
European calcified tissue society PhD training course
2014 Workshop on Photoshop and Illustrator
2015 Biomedical English Writing course

(Inter)national Conferences

2012 The Netherlands Institute for Regenerative Medicine Meeting, Amsterdam
Annual Meeting of the Dutch Society of Calcium and Bone, Zeist
2013 Internal Medicine Science Days, Antwerp, Belgium
Molecular Medicine Day, Rotterdam
Interbone Annual Meeting, Lisbon, Portugal (oral presentation)
International Conference of Children's Bone Health
The Netherlands Institute for Regenerative Medicine Consortium Meeting, Utrecht
2014 Internal Medicine Science Days, Antwerp, Belgium (poster presentation)
Molecular Medicine Day, Rotterdam (poster presentation)
International Society for Extracellular Vesicles Meeting, Rotterdam
European Calcified Tissue Society Meeting, Prague, Czech Republic
Interbone Annual Meeting, Prague, Czech Republic (oral presentation)
Dutch Society for Stem Cell Research Meeting, Groningen (poster presentation)
Matrix Biology Europe Meeting, Rotterdam (poster presentation)

The Netherlands Institute for Regenerative Medicine Consortium Meeting, Rotterdam (poster presentation)
Annual Meeting of the Dutch Society of Calcium and Bone, Zeist (oral presentation)
Annual Meeting of Dutch Society for Biomaterials and Tissue Engineering, Lunteren (oral presentation)

2015 Internal Medicine Science Days, Antwerp, Belgium (oral presentation)
Molecular Medicine Day, Rotterdam (poster presentation)
Dutch Society for Stem Cell Research Meeting, Utrecht (poster presentation)
European Calcified Tissue Society Meeting, Rotterdam (poster presentation)
Annual Meeting of the Dutch Society of Calcium and Bone, Zeist (oral presentation)
Annual Meeting of Dutch Society for Biomaterials and Tissue Engineering, Lunteren (oral presentation)

2016 Internal Medicine Science Days, Antwerp, Belgium (oral presentation)
European Calcified Tissue Society Meeting, Rome (poster presentation)
Annual Meeting of the Dutch Society of Calcium and Bone, Zeist (oral presentation)

Teaching Activities

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2016 Yik-Yang Kan, Hogeschool, Rotterdam

Awards

2013 *Erasmus Trustfond Travel Grant* for internship within Interbone Marie Curie Fellowship program at UNESP, SP, Brazil
2015 *Best poster award* Molecular Medicine Postgraduate School Day

Other activities

Weekly endocrinology lectures (2012-2016)
Weekly Skeleton Research Meetings (2014-2016)
Organization of the International Conference of Childen's Bone Health Meeting (2013)
Organization of the Internal Medicine Labday (2014)
Chairing Session at Annual Meeting of the Dutch Society of Calcium and Bone (2015)

PUBLICATIONS

Related to this thesis

Baroncelli M, Drabek K, Eijken M, van der Eerden BCJ, van de Peppel HJ, van Leeuwen JP. A single two-day-pulse of Activin-A leads to a transient change in osteoblast gene expression and reduction in extracellular matrix mineralization. *Submitted*

Baroncelli M, Fuhler GM, van de Peppel HJ, Zambuzzi WF, van Leeuwen JP, van der Eerden B, Peppelembosch MP. Human mesenchymal stromal cells in adhesion to cell-derived extracellular matrix and titanium: comparative kinome profile analysis. *Submitted*

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Previous publications

Morhayim J, **Baroncelli M**, van Leeuwen JP. Extracellular Vesicle: specialized bone messengers. Arch Biochem. Biophys. 2014 Nov 1;561:38-45. doi:10.1016/j.abb.2014.05.011 *Review*

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ACKNOWLEDGEMENTS

'Life is like riding a bicycle, to keep your balance, you must keep moving'
Albert Einstein

Finally, we have come to the last part of my thesis, the most important for me. Since I started my PhD, I have never believed I could make it to the end, for this I would like to acknowledge those of you who made the realization of this thesis possible and helped me directly or indirectly during this journey toward my PhD. I have to admit that before coming to Rotterdam I barely had an idea of where Rotterdam was, but now, after so many years, I really consider Rotterdam a second home (and I have learnt where it is). It has been an amazing adventure, I met so many people that helped me grow both as researcher and as a person and supported me during this time. I am incredibly grateful for this.

The first words of gratitude are for my promotor **Prof. Johannes van Leeuwen.** Dear **Hans,** thank you for giving me the opportunity to be part of the Bone Lab. Your curiosity and enthusiasm have been a continuous source of inspiration and motivation for me. Thank you for always reminding us to 'be like the osteocytes' and connect with people. I learnt a lot from you, from your broad and deep knowledge and I am sincerely grateful for your constructive comments during our meetings and your guidance.

I am very grateful to my co-promotor **Dr. Jeroen van de Peppel.** Dear **Jeroen,** thank you for your daily supervision and patience through these years. I still remember when you told me that even when you had to move to another office, you would still be there and I could still bother you. You have always been answering my questions and, indirectly, I have learnt from you how to become independent. Thank you for your constructive comments during our discussions, for letting me learn from your expertise on data analysis, and for the translation of the summary, but especially for letting me discover some enlightening Excel tricks, for the skype calls when I was in Brazil, the awesome photo album and the everyday chats in the office.

I would like to acknowledge the members of my reading committee, **Prof. Gerjo van Osch, Prof. Leo Hofland** and **Prof. Jan de Boer**, thank you for the time you invested in reading my dissertation. Dear **Gerjo**, you are a very inspiring scientist and I learnt a lot from your broad knowledge during the 'Skeleton meetings'. I thank you also for our discussions about the future, work-life balance and women in science. Dear **Leo**, I am sincerely grateful for all the interesting discussions during the Thursday Endo meetings. Dear **Jan**, I truly appreciated the insightful discussion we had at the NBTE meetings that could improve my project.

I would like to express my gratitude to **Prof. Hans van Leeuwen, Dr. Jeroen van de Peppel, Prof. Jan de Boer, Prof. Leo Hofland, Dr. Luc van der Laan, Prof. Peter ten Dijke, Prof. Maikel Peppelenbosch, Prof. Peter van der Spek** and **Prof. Eppo Wolvius**, I am honored that you accepted to be part of the plenary doctoral committee and invested your time to be present at my defence.

During these years I had the honor and the luck to meet many scientists who helped me reach this point, and I am very grateful for this. I would like to acknowledge **Prof. Jeroen Demmers** for all the help with the proteomics data, and **Enrique Rull-Trinidad, Dr. Luigi Sasso, Dr. Hugo Perez-Garza** and **Prof. Murali Ghatkesar** for your support with the AFM microscope and data analysis. I would like to extend my gratitude to **Dr. Lidy Fratila-Apachitei** and **Ingmar van Hengel** for finally letting me visualize my ECM, your help with SEM and image analysis was precious, I truly appreciate that.

I am very grateful to **Dr. Gwenny Fuhler**, **Prof. Maikel Peppelenbosh,** and **Prof. Willian Zambuzzi**. Dear **Gwenny** and **Maikel**, thank you for your help in introducing me to the 'kinome profiling world' with so much patience, for your help and insightful advices during the experiments and the manuscript writing. Dear **Willian**, I hope that our collaboration will soon come to a nice publication. Thank you for all your support during my staying in Brazil, for making it a great experience and for your time and fruitful discussions.

A big thought towards **Prof. Theo Visser**, whose recent loss was deeply felt by everybody. I feel extremely honored and grateful for the insightful discussions at the Thursday endo meetings and for the nice chats at the Science days. Your curiosity and love for science will continue to inspire.

My big gratitude to my "Paraninfen", **Marialuisa** and **Jess**. I could never thank you enough for your support and great help to organize the thesis and the defence. Dear **Marialuisa, Isa** bellina, non riesco davvero ad immaginare come avrei fatto senza di te a Rotterdam, c'eri sempre per me, nei momenti brutti e in quelli belli. Una presenza costante fin dai primi momenti, che si è poi evoluta in una delle più belle e forti amicizie che ho, sei una sorella acquisita per me. Sarà bellissimo poterti stare accanto in uno dei giorni più belli della tua vita, grazie per questo. Sono così contenta per te e Andrea, i miei riferimenti di Rotterdam, e sono così felice che possiamo mantenere la nostra amicizia anche a 1500 km di distanza. Dear **Jess**, I am so grateful we had the chance to share the office, thank you for your support and help in the lab, but also for your friendship outside the Bone lab. I was really lucky to have you as an example of smart PhD student, I have been really admiring how you successfully combine work and personal life. I have always been impressed by how you could make everything look so simple and feasible. I wish you all the best to you, Hugo and Mateo with the upcoming baby!

During these years I spent quite some time in the lab, and I had the honor and the luck to share it with many people, who not only helped me reach this point with their encouraging attitude, but also created a nice working environment, during and after work hours. I express my sincere gratitude to the former and the current members of the **Bone lab,** Andrea, Annelies, Bram, Cindy, Iris, Jeroen, Joëlle, Ksenija, Marijke, Marjolein, Rodrigo, Shourouk, Shuang, Tanja, Yiman, Yuan, and the Arcarios group, Anke, Bianca, Marc, Marco. In this moment I realize how many we have been through the years. Words of gratitude go also to the exchange students that came through the lab, Adriana, Andreas, Aranka, Jyoti, Natasja, Pavitra, Pui, Tim, and especially to my students Siddharth, Shevarna and Yik, thank you for all the work you did, I truly appreciated it, good luck for your future studies/jobs. Dear **Bram**, my additional co-promotor. I really do not know how to thank you for all the help, support and your time, during my stay in the lab but also during the last months when I was not in Rotterdam, you were always there with your continuous encouragement. I truly appreciated the discussions we had about research, and not directly related to research, especially at the ECTS cocktails. I guess it was also for your nice words about me if I got my current position in Ola's lab, I express my gratitude also for this. I believe and hope we can carry on with our collaboration to bring bone and growth plate even closer. Dear **Marjolein**, I am sincerely grateful for your continuous support and encouragement. I am glad we shared our birthdays and I am sure we will carry on with the tradition with nice celebrations and cake. Dear **Andrea**, you and Jess were both my inspiration on how to be a great PhD student, you were always present to ask me how it was going, it was so nice sharing these years with you in the lab and outside of it. I wish you all the best to you and Teun for your new baby. Dear **Tanja**, I still remember the picture I had of you before actually meeting you: the queen of the wedding celebrations. Thank you so much for your help with the cells, but especially for your friendship outside the lab, you are such a nice person and I am really glad I met you. I am so proud of you having made and still making tiramisu for your anniversary! Dear **Cindy**, thank you for your help and support and especially your friendship in and outside the lab, that became stronger and stronger. I am truly grateful we can still maintain it, though with more km in between. Dear **Marijke**, I cannot thank you enough, for all your patience and help with all my questions and experiments. Whenever I felt lost, I always knew where to find you! You are really the backbone of the bone lab. Dear **Iris**, thank you for the time in the lab, the pre-cultures, the nice chats in the culture room, and especially for turning the music on in the lab! Dear **Annelies**, you have been so helpful with me even before I came, you were always there to help out and so amazingly efficient, thank you. Dear **Joëlle,** thank you for the nice chats in the office, I wish you best of luck for your PhD. Dear **Rodrigo**, you left before I started, but thank you for starting the work on the

extracellular matrix and inspiring its development. I want to express my gratitude especially for your thesis book, or books I should say, as I read it so many times that the first copy was wore out. Dear **Marco** and **Ksenija**, I am sincerely grateful that you successfully pioneer the Activin-A work, to which I could contributed in a small part.

My words of gratitude go also the Orthopedic Lab at the 16th floor. Dear **Gerjo, Roberto, Eric** and **Ivonne**, I am very grateful for your insightful discussion during the 'Skeleton meetings', NBTE and Molmed days. Dear **Andrea, Callie, Caoimhe, Johannes, Kavitha, Lizette, Maarten, Panithi, Shorouk, Simone, Sohrab, Wu,** thank you for 'adopting' me and making me feel another PhD student of the crew when I was the only one left at the 5th floor, your help at work, and especially the drinks at Boudewijn (and not only) after work.

My gratitude goes also to the people I met through the corridors of the 5th floor through these years, especially **Carolina, Fjorda, Jeroen, Kate, Martin, Marije, Sabine, Sanne, Thomas, Vincent.** Dear **Carolina**, amiga, I am very thankful for your friendship, coffees and cakes, and thank you so much for the nice time we had at ECTS and for introducing me to the first person I met in Stockholm, Alice. Dear **Marije**, thank you for introducing me to 'our' island, for our nice dinners and drinks. I wish you all the best for you and Berrie, you totally deserve it. Dear **Martin**, thank you for the nice chats in the culture room, and the gelato I "had to" taste every time. Good luck for your crazy trip, I am waiting for awesome pics!

Apparently, Rotterdam was not north enough and I decided to move further north. These last months have been quite challenging, and I am very grateful for my current colleagues of the **Growth and Cartilage Biology group**. Dear Ola, thank you for letting me join your group, I truly appreciated your trust and understanding, and I am sincerely grateful for your patience during these months. You are so encouraging with people, I really admire this. Dear Alexandra, Ameya, Lars, Sigrun and Zelong, thank you for making me feel welcome in Stockholm and in the lab and helping me every day. Alice, sei stata gentilissima da prima che accettassi di venire a Stoccolma. Grazie per i nostri pranzi, caffè, uscite e chiacchierate estremamente necessari! Ti ringrazio per avermi accolto al CMM e a Stoccolma, cominciando a farmela sentire un po' più la mia terza casa.

Un ringraziamento immenso a tutti gli amici italiani che ho conosciuto nei vari anni, my "Italian mafia" as Jeroen was always saying. Siete stati di fondamentale importanza per sopravvivere a questa città fredda, grigia e ventosa, ma poi siete diventati come una seconda famiglia. Mi sento molto fortunata ad avervi incontrato. Agli Italians che mi hanno accolta e fatta sentire a casa fin dai primi giorni a Rotterdam: **Giulia A, Giulia P, Claudia**, **Paolo, Federichino, Franceschino, zia Silvana, Francesca**, **Carmen** e **Gianluca, Antonio** e **Stefania, Valerio** e **Valentina, Pier** e **Laura**, **Sara** e **Stefan, Davide G, Eva, Marco, Matteo** grazie per avermi accolta all'inizio.

Giulia A, piccola sore, sei andata via dopo poco da Rotterdam, ma c'eri e ci sei sempre. Giulia P, bellina, grazie per tutte le fantastiche serate in giro per Rdam, c'era sempre una buona occasione per festeggiare. Claudia, senza di te sarei stata persa all'inizio, ricordo ancora la cena al Western dopo il mio colloquio. Paolo, qua a Stoccolma non ho neanche un forno, ma un piatto da forno sarebbe fondamentale per pane e stracchino. Federichino, se sono arrivata fino a qua mi sa che ce l'ho fatta a sopravvivere all'uomo nero, grazie per il tuo supporto quando eri in the office, ma anche dalla cucina del 5 piano quando sei tornato al neuroendo, ed avermi fatto essere la tua superparaninfa. Franceschino, mi ricordo ancora quando avevamo caricato le scatole del trasloco sulla bici e attraversato il ponte. Invece che venire in Svizzera mi sono allontanata ancora di più, ma ci teniamo in contatto e vi aspetto a Stoccolma. Zietta, grazie per tutti i nostri tè e tisane con Isa, e tutte le chiacchierate ai piani altissimi dell´MC. Francesca, sei tornata in Italy prima che io arrivassi a Rotterdam, ma la tua presenza si sentiva anche dopo, così come le tue feste memorabili. Sara e Stefan, Davide G, Eva e Marco e Matteo, come dimenticare tutte le prelibatissime cene nella fantastica cucina di Davide. Agli Italians incontrati via via **Albachiara, Ottavia, Francesca V, Francesca B, Alessandro, Andrea, Peppe** che hanno contribuito a rendere le serate a Witte e dintorni speciali. Dimenticherò sicuramente qualcuno e lo realizzerò appena avrò stampato tutto. Un ringraziamento speciale a Rotterdam, perché mi ha fatto incontrare voi **Simona, Celia, Isa, Andrea, Luca, Andrea, Guido**, **Francesco, Anita, Ilario, Marco, Anna, Davide, Alberto**. Alcuni di voi hanno cambiato città (me compresa), ma posso dire che siete stati e siete fondamentali per me in questi anni, siete davvero stati la mia seconda famiglia. Andrea, ti devo ringraziare per tutte le cose superbuonissime che hai cucinato, e visto che a Stoccolma ho una cucina un po' finta dove non posso farti cucinare, la prendo come scusa per invitarmi da voi e le gattine. Luca, sei l'unica persona in grado di apprezzare l'inverno svedese, ti aspetto per un corso accelerato di positività. Andrea, grazie per tutti i caffè da Albert, cene e birrette di fondamentale importanza per sopravvivere all'olandesità generale e al luogo di gioia. Guidooo, i tuoi consigli filosofici sono sempre con me anche a Stoccolma, anche se non sembra, ti ascolto, a volte. Francesco, ce la faremo ad organizzare la gita a Montale, non temere, so che non vedi l'ora. Vedete di non cambiare casa con il tuo coinquilino che Schiekade 107 con il fantastico giardino è uno dei riferimenti di Rotterdam. Anita, non smetterò mai di ringraziarti per la bellissima (almeno per me) copertina di questa tesi, che pazienza hai avuto. Ila, dopo essere sopravvissuti al ferragosto a Rotterdam, i prossimi solo mare. Marco e Anna, lo so che Stoccolma è troppo a nord, quindi verrò io in gita da voi, magari a Napoli. Zio Davide, sono così contenta che ti sei preso un po' di cose della mia casetta, ti aspetto a Stoccolma, mi raccomando. Alberto, ogni tanto dobbiamo organizzare un weekend a Rdam come la sorpresa tua e di Simo, così per vedere i nostri amici di

Witte come stanno. **Simona** e **Celia**, non ringrazierò mai abbastanza Rotterdam e l'Erasmus per averci fatto incontrare. Non servono molte parole per dirvi quanto vi voglio bene, siete le altre mie sorelle acquisite. Come mi avete detto voi, le persone che rendono speciali le pagine della vita, saranno sempre presenti. **Mary** e **Mau** siete sempre con me anche se non ci sentiamo spesso, ma ogni volta che torno mi fate sempre sentire come se non fossi mai partita. **Marghe, Ire, Simo** che porto con me dall'università, siete un continuo supporto e fonte di ispirazione.

Zia Rita, come direbbe lo zio Aldo, ora hai anche questa tesi da leggere, basta con le ripetizioni di inglese! Nonno Pietro, penso saresti orgoglioso. Grazie a tutti i parenti vari, non vi nomino uno ad uno che siamo troppi, è un sacco bello quando ci ritroviamo.

Il ringraziamento più grande va alla mia famiglia, mamma, babbo, Angy, Ale, Chiara, Ste, Anna, Lapo e Nhoa. Le parole non riescono ad esprimere i sentimenti al meglio, soprattutto le mie per voi. Vi voglio bene. Senza il vostro supporto incondizionato e affetto semplicemente tutto questo non sarebbe stato possibile (e Ste non avrebbe potuto fare due traslochi e attraversare quattro nazioni in un giorno). Senza di voi non ce l'avrei fatta e non sarei la persona che sono. Non mi avete fatto mai sentire lontano da casa, siete sempre stati i primi al mio fianco ogni volta che decidevo di partire per una nuova avventura. Siete sempre con me.

Marta