
Contents

<i>Preface</i>	<i>v</i>
<i>Contributors</i>	<i>xi</i>
PART I MURINE SKELETAL DEVELOPMENT, REPAIR, AND GENETIC MODELS	
1 Overview of Skeletal Development	3
<i>Tatsuya Kobayashi and Henry M. Kronenberg</i>	
2 Overview of Skeletal Repair (Fracture Healing and Its Assessment)	17
<i>Elise F. Morgan, Anthony De Giacomo, and Louis C. Gerstenfeld</i>	
3 Advantages and Limitations of Cre Mouse Lines Used in Skeletal Research	39
<i>Florent Elefteriou and Greig Couasnay</i>	
PART II SKELETAL REPAIR, PARABIOSIS, TRANSPLANTATIONS, AND ORGAN CULTURES	
4 Generation of Closed Transverse Fractures in Small Animals	63
<i>Anthony De Giacomo, Elise F. Morgan, and Louis C. Gerstenfeld</i>	
5 A Mouse Femoral Osteotomy Model to Assess Bone Graft Substitutes	75
<i>Ryan P. Trombetta, Emma K. Knapp, and Hani A. Awad</i>	
6 Surgical Induction of Posttraumatic Osteoarthritis in the Mouse	91
<i>Robert D. Maynard, David A. Villani, William G. Schroeder, Douglas J. Adams, and Michael J. Zuscik</i>	
7 Parabiosis: Assessing the Effects of Circulating Cells and Factors on the Skeleton	105
<i>Benjamin Alman and Gurpreet Baht</i>	
8 Murine Limb Bud Organ Cultures for Studying Musculoskeletal Development	115
<i>Martin Arostegui and T. Michael Underhill</i>	
9 Murine Limb Explant Cultures to Assess Cartilage Development	139
<i>Manuela Wuelling and Andrea Vortkamp</i>	
10 Renal Capsule Transplantation to Assay Angiogenesis in Skeletal Development and Repair	151
<i>Anais Julien, Simon Perrin, Rana Abou-Khalil, and Céline Colnot</i>	
PART III STRUCTURAL, HISTOLOGICAL, AND MOLECULAR ANALYSES ON SKELETAL TISSUES AND TISSUE SECTIONS	
11 MicroCT for Scanning and Analysis of Mouse Bones	169
<i>Yung Kim, Michael D. Brodt, Simon Y. Tang, and Matthew J. Silva</i>	

12	Four-Point Bending Testing for Mechanical Assessment of Mouse Bone Structural Properties	199
	<i>Hattie C. Cutcliffe and Louis E. DeFrate</i>	
13	Phenotyping Intact Mouse Bones Using Bone CLARITY	217
	<i>Jennifer B. Treweek, Aidan Beres, Nathan Johnson, and Alon Greenbaum</i>	
14	Processing and Sectioning Undecalcified Murine Bone Specimens	231
	<i>Thomas B. Bemenderfer, Jonathan S. Harris, Keith W. Condon, Jiliang Li, and Melissa A. Kacena</i>	
15	Preparation of Thin Frozen Sections from Nonfixed and Undecalcified Hard Tissues Using Kawamoto’s Film Method (2020)	259
	<i>Tadafumi Kawamoto and Komei Kawamoto</i>	
16	Demineralized Murine Skeletal Histology	283
	<i>Anthony J. Mirando and Matthew J. Hilton</i>	
17	Assessment of Osteocytes: Techniques for Studying Morphological and Molecular Changes Associated with Perilacunar/Canalicular Remodeling of the Bone Matrix	303
	<i>Neba S. Dole, Cristal S. Yee, Charles A. Schurman, Sarah L. Dallas, and Tamara Alliston</i>	
18	Cell Lineage Tracing: Colocalization of Cell Lineage Markers with a Fluorescent Reporter	325
	<i>Yan Jing, Patricia Simmer, and Jian Q. Feng</i>	
19	Immunofluorescent Staining of Adult Murine Paraffin-Embedded Skeletal Tissue	337
	<i>Neta Felsenthal and Elazar Zelzer</i>	
20	Detection of Hypoxic Regions in the Bone Microenvironment	345
	<i>Wendi Guo and Colleen Wu</i>	
21	EdU-Based Assay of Cell Proliferation and Stem Cell Quiescence in Skeletal Tissue Sections	357
	<i>Marco Angelozzi, Charles R. de Charleroy, and Véronique Lefebvre</i>	
22	Whole Mount In Situ Hybridization in Murine Tissues	367
	<i>Deepika Sharma, Matthew J. Hilton, and Courtney M. Karner</i>	
PART IV PRIMARY CELL ISOLATIONS, CULTURES, AND ASSAYS		
23	Bone Marrow Stromal Cell Assays: In Vitro and In Vivo	379
	<i>Pamela G. Robey, Sergei A. Kuznetsov, Paolo Bianco, and Mara Riminucci</i>	
24	Isolation and Culture of Periosteum-Derived Progenitor Cells from Mice	397
	<i>Chinedu C. Ude, Girdhar G. Sharma, Jie Shen, and Regis J. O’Keefe</i>	
25	Isolation and Culture of Murine Primary Chondrocytes: Costal and Growth Plate Cartilage	415
	<i>Yiban Liao, Jason T. Long, Christopher J. R. Gallo, Anthony J. Mirando, and Matthew J. Hilton</i>	

26 Isolation and Culture of Neonatal Mouse Calvarial Osteoblasts..... 425
*Madison L. Doolittle, Cheryl L. Ackert-Bicknell,
and Jennifer H. Jonason*

27 Mitochondrial Function and Metabolism of Cultured Skeletal Cells..... 437
Li Tian, Clifford J. Rosen, and Anyonya R. Guntur

28 Radiolabeled Amino Acid Uptake Assays in Primary Bone
Cells and Bone Explants 449
Leyao Shen and Courtney M. Karner

29 RANKL-Based Osteoclastogenic Assay from Murine Bone
Marrow Cells..... 457
Zhenqiang Yao, Lianping Xing, and Brendan F. Boyce

30 Hematopoietic Stem Cell Cultures and Assays..... 467
Benjamin J. Frisch

Index 479