
Contents

1 Stem Cell and Its Niche	1
Terminology of Stem Cell Niche	1
First Hypothesis and Evidence of Stem Cell Niche.....	1
Role of Extracellular Matrix in Regulating Stem Cell Niche	2
Subchapter A.....	2
Stem Cell Niche in the Lung.....	2
Introduction.....	2
Evidence of Lung Stem Cells	3
Evidence from Ex Vivo Studies	3
Rat Tracheal Xenograft Model.....	3
Injury by Naphthalene.....	4
Response to Naphthalene Injury to Lung.....	4
Tracheal Injury Model by Inhaled Sulfur Dioxide.....	5
Repair After Damage by Inhaled Oxidants.....	5
Lung Stem Cell Niche: Potential	5
Scope of Further Research	5
Subchapter B.....	6
Other Stem Cell Niches	6
Potential of Cellular Regeneration from Hair	
Follicle Stem Cells.....	6
Isolation of Hair Follicle Stem Cells: Procedure	6
Expression of Various Stem Cell Markers	7
Differentiation of Stem Cells to Different	
Lineage-Specific Cells	7
Differentiation to Melanocytic Cells.....	7
Differentiation to Neural Cells.....	7
Smooth Muscle Cell Differentiation of Human	
Hair Follicle Cells	7
Subchapter C.....	8
Mesenchymal Stem Cells.....	8
Brief History	8
Morphology.....	8
Characteristics.....	9
Isolation of MSC from Human Umbilical Cord	9
Immunophenotyping of Human Umbilical Cord	
Blood MSCs.....	9

BrdU Incorporation and Proliferation Assay	9
RT-PCR Analysis	9
Transfection of UC-MSCs with cDNA and mRNA.....	10
Clinical Significance of MSC	10
Capacity to Migrate and Engraft.....	10
Neuronal Differentiation of MSCs.....	10
Immunomodulatory Functions of MSCs	10
Secreting Multiple Bioactive Molecules.....	11
Future Directions	11
References.....	12
2 A Mini Review on Stem Cells, Their Study Models, and Effect of Trace Elements	15
Introduction.....	15
Classification of Stem Cells Based on Their Differentiation Potential.....	16
Classification of Stem Cells Based on Origin and Their Sources.....	17
Embryonic Stem Cells (ESCs).....	17
Fetal Stem Cells (FSCs).....	17
Perinatal Stem Cells.....	17
Adult Stem Cells (ASCs).....	18
iPS Cells.....	18
Pathway of Stem Cell Activity.....	18
Stem Cell Niche.....	18
Stem Cell Mobilization and Homing.....	19
Stem Cell Differentiation and Plasticity	19
Neural Stem Cells as a Model for Stem Cell Development.....	20
Stem Cell Engraftment.....	20
Model of Stem Cell Research	20
<i>Drosophila</i> <i>Melanogaster</i>	20
Conclusion	22
Effect of Lithium: A Trace Element on Stem Cells	22
References.....	23
3 Use of Stem Cells in Drug Screening.....	25
Introduction.....	25
Stem Cells in Drug Toxicity Screening.....	25
Stem Cells as Screen for Differential Toxicity	27
Cell Lines Used in Drug Screening and Toxicity Studies.....	27
Clinical Candidate Optimization.....	27
Intestinal Absorption.....	30
Drug Metabolism	31
Toxicology	32
Stem Cells and ADMET	32
Conclusion	35
<i>Drosophila</i> as a Model for Studies on Stem Cells	37
References.....	37

4 Tissue Differentiation of ESC into Lung Cells and Functional Validation	39
Introduction.....	39
Results.....	40
Differentiation of hES Cells Is Accompanied by Sequential Downregulation of Pluripotent Markers	40
Variation in Growth Media Skews Differentiation of hES Cells to AEI Cell, AEII Cell, and Clara Cell Phenotypes	41
Inhibition of Wnt/ β -Catenin/CBP Signaling Promotes Differentiation of hES Cells to AEI Cell Phenotype.....	41
Clonogenic Potential of hES Cells Declines with Differentiation.....	47
Engraftment of Differentiated hES Cell Transplants in Mice with Lung Fibrosis.....	48
Differentiated hES Cell Transplants Reduce Pulmonary Inflammation and Fibrosis Induced by Bleomycin.....	49
Differentiated hES Cell Transplants Increase Airway Epithelial Cells and Progenitors in Mice with Pulmonary Fibrosis	50
Discussion	52
Materials and Methods.....	56
Ethics Statement.....	56
Expansion of H7 hES Cells.....	56
Embryoid Body Formation	56
Generation of Non-ciliated Pulmonary Epithelial Cells	57
Phenotypic Analysis of Cells	57
Cell Viability	58
Clonogenic Growth of Cells Derived from hES Cells	58
Mouse Model of Pulmonary Fibrosis and Transplantation of Differentiated H7 hES Cells	58
Analysis of Colony-Forming Units in Tissue Compartments.....	59
RNA Isolation	60
qPCR Analysis	60
Analysis of Collagen Content in Lung.....	60
Detection of Human Cells in Mouse Lung	60
Immunohistochemistry	62
Microscopy	62
Transmission Electron Microscopy.....	62
Statistical Analysis	62
Background.....	62
Methodology and Principal Findings.....	63
Conclusions.....	63
References.....	63

5	Validation of Lung Stem Cell Niche	67
	Introduction.....	67
	Materials and Methods.....	68
	Mice	68
	Mouse Model of Bleomycin-Induced Pulmonary Fibrosis.....	68
	BrdU Pulse-Chase Assay	68
	BALf	69
	Lung Parenchyma	69
	Lung Histology	69
	Fluorescence-Activated Cell Sorter (FACS) Analysis	69
	CFU-c Assay	69
	Statistical Analysis	70
	Results.....	70
	Bleomycin-Induced Idiopathic Pulmonary Fibrosis Model.....	70
	Assessment of Extent of Fibrotic and Inflammatory Damage in the Lung Post-bleomycin Treatment	70
	Inflammatory Cell Accumulation in the Lungs and Airways Over Time Post-bleomycin Treatment.....	70
	Stem Cell Niche Characterization in Bleomycin-Induced Injury Model	72
	Rationale of Long-Term Assay	72
	Long-Term BrdU Pulse-Chase Assay	73
	Detection of Stem Cells by CFU-C	73
	Detection of Stem Cells by Marker Expression.....	73
	Characterization of BrdU+ Cells	73
	Characterization of Non-stem Cells (BrdU-).....	75
	Short-Term Hoechst SP Experiment	75
	Rationale of Short-Term Assay	75
	Analysis of Inflammatory Cells Post-bleomycin Treatment in Single and Double Knockout Mice	76
	SP Cells Isolated from Pre- and Post-bleomycin Treatment (Day 7).....	76
	Cells in BALf and Lung Post-bleomycin Over Time.....	76
	Characterization of Sorted SP Cells in Ex Vivo Culture.....	77
	Discussion	77
	Conclusion	83
	References.....	83
6	Model Organisms in Science and Research	85
	What Is Model Organism?.....	85
	<i>Caenorhabditis elegans</i> as a Model Organism	85
	<i>C. elegans</i> Timeline.....	86
	Major Areas of Study	86
	Lineage Studies	86
	Apoptosis	86
	Aging.....	88
	Nervous System	88
	Gene Expression and Functional Studies.....	88

<i>C. elegans</i> Model Identifies Genetic Modifiers of α -Synuclein Inclusion Formation During Aging	88
<i>Caenorhabditis elegans</i> Muscle: A Genetic and Molecular Model for Protein Interactions in the Heart	88
<i>Caenorhabditis elegans</i> : A Model Organism for Investigating Immunity	88
Validated <i>C. elegans</i> Disease Models	89
The <i>Drosophila</i> Story.....	89
Salient Features of This Model	90
Areas of Study.....	90
Zebra Fish as a Model Organism	90
Disease Areas of Study Using Zebra Fish as a Model	90
<i>Xenopus</i> as Model Organism	91
Introduction.....	91
Advantages.....	91
Causes	92
Immunological Aspects of Using <i>Xenopus laevis</i> as a Model Organism	92
Skin Graft Rejection in Adult <i>X. laevis</i>	92
Immune System of <i>X. laevis</i>	92
<i>X. laevis</i> Is Interestingly Getting Replaced by <i>X. tropicalis</i>	93
Causes	93
Conclusion	93
Chick as a Model System (Sc. Name, <i>Gallus gallus</i>).....	94
Popular Experimental Manipulation	94
Role of Chick in Immunological Studies.....	94
Immunological Mouse Models	96
Applications	96
Advantages.....	96
Disadvantages	96
Background on Mouse as a Model Organism.....	97
Mouse Models for Addiction	97
Applications	97
Advantages.....	98
Disadvantages	98
Applications	99
Characteristics.....	99
Applications	99
Characteristics.....	100
Features of NSG Mouse.....	100
Applications	100
Applications	101
Applications	101
Applications	101
Concluding Remarks.....	104
References.....	104
Suggested Reading.....	104

7	Models for Studies in Regenerative Medicine	105
	What Is the Need for Model Organisms for Studying Nuances of Regenerative Medicine?.....	105
	Advantages (Pros) of Using Models	106
	Disadvantages (Cons) of Using Models.....	106
	Concluding Remarks.....	106
	Why We Need Model Organisms in Regenerative Research Studies?	107
	Do All Animals Regenerate?.....	107
	Need for Model Systems.....	107
	Achieved so Far.....	107
	<i>Planaria (Schmidtea mediterranea)</i>	109
	Cell Culture from Marine Invertebrates: New Insights for Capturing Endless Stemness	109
	Planarians (<i>Schmidtea mediterranea</i>)	109
	Regeneration in Planaria	109
	The Process	111
	Signaling	111
	Zebra Fish (<i>Danio rerio</i>).....	111
	Regeneration in Zebra Fish.....	111
	African Clawed Frog (<i>Xenopus</i> sp.).....	112
	Regeneration in <i>Xenopus</i>	112
	Hydra (<i>Hydra vulgaris</i>).....	112
	Regeneration in <i>Hydra</i>	112
	The Process	113
	Mice Models	113
	Regeneration in Mice	113
	Conclusion	113
	Future Strategy.....	114
8	Role of Progenitors in Pulmonary Fibrosis and Asthma	115
	Introduction.....	115
	Tissue Repair in Lung Disorder	116
	Idiopathic Pulmonary Fibrosis (IPF), Progenitors, and Niche Plasticity	116
	Asthma, Progenitors, and Niche Plasticity	117
	Bone Marrow Cell Mobilization in Response to Injury.....	118
	Migration to the Site of Injury	118
	Implication of Fibrocytes as Common Denominator for Remodeling	119
	Fibrocytes in the Lumen of the Airway	119
	Conclusion	119
	References.....	119
9	Some Concepts in Studies of Kidney Regeneration	123
	Kidney Regeneration.....	123
	Introduction.....	123
	Disease Models on Renal Ischemia and Reperfusion	124

Regenerative Medicine in Acute and Chronic Renal Complication	125
Future Perspectives: The Way Forward	126
Kidney Development	126
Molecular Basis of Kidney Development	126
Concepts on Renal Stem Cell Niche and iPSC	127
The Stem Cell Niche as an Entity of Action	127
The Renal Papilla Is a Niche for Adult Kidney Stem Cells	128
Renal Capsule as a Stem Cell Niche	128
Induced Pluripotent Stem Cell (iPSC)	129
Generation of iPSC from Human Renal Proximal Tubular Cells with Only Two Transcription Factors: OCT4 and SOX2	129
Kidney Side Populations and Their Roles	131
Role of Stem Cells in Repair of AKI	131
To Test That, Native Kidney Cells That Reside in A Niche in the Kidney Provide Robust Contribution to the Repair of Kidney Tubules Following Injury	132
To Evaluate the Differential Contribution of Extrarenal Cells and Intrarenal Cells to Renal Repair	132
Selection of the Optimal Cell for Kidney Regeneration: Fetal, Adult, or Reprogrammed Stem Cells	132
Self Assembly of Cultured Cell Populations into Integral Tissue in Context of Kidney Regeneration	133
Whole-Organ Tissue Engineering: Decellularization and Recellularization of Three-Dimensional Matrix Scaffolds	138
The Nanotechnology Approach	140
Developing Kidney-Like Tissue from Unbranched Epithelial Structure	140
The Group of Rosine et al. in the Year 2010 Again Published Their Work on Engineering Kidney Tissues In Vitro from Cultured Cells	141
De Novo Kidney Regeneration: Bench to Bedside Approach	141
References	144
10 Ethics in Stem Cell Research	147
Opening Lines	147
Is Destroying Human Embryo Morally Ethical?	147
Does iPSC Eliminate All the Setbacks Associated with hES Cell Research?	147
Specific Principles Related to Stem Cell Research	147
Scenario and Regulation of Stem Cell Research in India	148
Categorization of SCR	148
Post Script	151



<http://www.springer.com/978-81-322-2052-7>

Perspectives in Regenerative Medicine

Ray Banerjee, E.

2014, XIX, 152 p. 79 illus., 44 illus. in color., Hardcover

ISBN: 978-81-322-2052-7