

Contents

List of Contributors *XI*

Preface *XV*

A Personal Foreword *XVII*

1	Introduction	<i>1</i>
	<i>Marianne Isabelle Martic-Kehl, Michael F.W. Festing, Carlos Alvarez, and P. August Schubiger</i>	
1.1	Animal Models in Biomedical Research	<i>1</i>
1.2	Animals in the Drug Development Process: Historic Background	<i>2</i>
1.3	Problems with Translation of Animal Data to the Clinic	<i>5</i>
1.4	Animal Studies in Anti-cancer Drug Development	<i>6</i>
1.5	Toward Relevant Animal Data	<i>7</i>
1.6	Aim of the Book	<i>8</i>
	References	<i>8</i>
 2	 Ethical Aspects of the Use of Animals in Translational Research	 <i>11</i>
	<i>Karin Blumer</i>	
2.1	Introduction	<i>11</i>
2.2	Today's R&D Environment	<i>11</i>
2.2.1	Four Emerging Trends Shaping Today's Debate	<i>13</i>
2.2.1.1	Growing Lack of Awareness of the Nature of Science and Research	<i>13</i>
2.2.1.2	Increased Pressure on Basic Research	<i>14</i>
2.2.1.3	Pressure to Assign "Special" Animals a Special Moral and Legal Status	<i>15</i>
2.2.1.4	A Reductionist Approach to the 3Rs	<i>16</i>
2.2.2	Preliminary Conclusions	<i>17</i>
2.3	"Do No Harm": the Essential Dilemma of Animal Research	<i>17</i>
2.4	Man and Animals in Philosophy: an Overview of Key Concepts	<i>18</i>
2.4.1	Anthropocentrism	<i>19</i>
2.4.2	Physiocentric Positions	<i>19</i>
2.4.2.1	Holistic Concepts	<i>19</i>
2.4.2.2	Radical Biocentrism	<i>20</i>

2.4.2.3	Pathocentrism	21
2.4.2.4	Moderate Biocentrism	22
2.5	Conclusions: Solving the Dilemma	23
	References	24
3	Study Design	27
	<i>Michael F.W. Festing</i>	
3.1	Introduction	27
3.2	Design Principles	28
3.3	Experimental Design	28
3.3.1	The Five Characteristics of a Well-Designed Experiment	29
3.3.2	The Determination of Sample Size	34
3.3.2.1	Power Analysis for the Determination of Sample Size	34
3.3.2.2	The Resource Equation Method of Determining Sample Size	36
3.3.3	Formal Experimental Designs	36
3.4	Conclusion	39
	References	39
4	Improving External Validity of Experimental Animal Data	41
	<i>S. Helene Richter, Chiara Spinello, and Simone Macri</i>	
4.1	Introduction	41
4.1.1	Individual Phenotype Is the Result of Genetic and Environmental Influences	41
4.1.2	Why Do Living Organisms Vary?	42
4.2	Variation in the Laboratory	43
4.2.1	How Is Inter-individual Variability Generally Dealt With?	43
4.2.1.1	Genetic Standardization	44
4.2.1.2	Environmental Standardization	44
4.2.1.3	Standardization of the Test Situation	46
4.3	The Fallacies	46
4.3.1	The Standardization Fallacy	46
4.3.2	The Developmental Match Fallacy	47
4.4	Future Perspectives: an Experimental Strategy Integrating Adaptive Plasticity and Fundamental Methodology	48
4.4.1	A Way Out of the Standardization Fallacy?	48
4.4.2	Favoring Adaptive Plasticity through the Provision of Test Strategies Matching Developmental Cues	53
	References	55
5	How to End Selective Reporting in Animal Research	61
	<i>Gerben ter Riet and Lex M. Bouter</i>	
5.1	Introduction	61
5.2	Definition and Different Manifestations of Reporting Bias	63
5.3	Magnitude of Reporting Biases	63
5.4	Consequences	65

5.4.1	Consequences of Reporting Bias in Human Randomized Trials	65
5.4.2	Consequences of Reporting Bias in Experimental Animal Research	66
5.5	Causes of Reporting Bias	66
5.6	Solutions	68
	References	73
6	A Comprehensive Overview of Mouse Models in Oncology	79
	<i>Divya Vats</i>	
6.1	Introduction	79
6.2	Xenograft Mouse Models	81
6.2.1	Cell-Line Xenograft Model	81
6.2.2	Patient-derived Xenografts	82
6.3	Genetically Engineered Mouse Models	83
6.3.1	Limitations	85
6.3.2	Chemical Carcinogenesis: N-ethyl-N-nitrosourea Mutagenesis	86
6.3.2.1	Alkylnitrosamide Compounds	86
6.3.3	Generation of a Transgenic Mouse Using Pronuclear Injections: Direct Insertion of DNA into Fertilized Zygote	87
6.3.4	Gene Targeting via Homologous Recombination in Embryonic Stem Cells: Gene Knockouts and Knock-Ins	87
6.3.5	Conditional Inactivation (or Activation) of Genes	89
6.3.6	Inducible Systems for Gene Targeting	90
6.3.7	RNA Interference for Gene Knockdown	92
6.4	Applications for GEMMs in Compound Development	93
6.4.1	Target Validation and Compound Testing	93
6.4.2	Chemoresistance and Toxicity	94
6.4.3	<i>In vivo</i> Imaging	94
6.5	Humanized Mouse Models: toward a More Predictive Preclinical Mouse Model	95
6.6	Conclusions: Potentials, Limitations, and Future Directions for Mouse Models in Cancer Drug Development	98
6.6.1	Potentials and Limitations	98
6.6.2	Future Directions	100
	References	101
7	Mouse Models of Advanced Spontaneous Metastasis for Experimental Therapeutics	109
	<i>Karla Parra, Irving Miramontes, Giulio Francia, and Robert S. Kerbel</i>	
7.1	Mouse Tumor Models in Cancer Research	109
7.2	The Evolution of Metronomic Chemotherapy	110
7.3	Development of Highly Aggressive and Spontaneously Metastatic Breast Cancer Models	112

7.4	Is There Any Evidence that Models of Advanced Metastatic Disease Have the Potential to Improve Predicting Future Outcomes of a Given Therapy in Patients? 113
7.5	Metronomic Chemotherapy Evaluation in Preclinical Metastasis Models 116
7.6	Experimental Therapeutics Using Metastatic Her-2 Positive Breast Cancer Xenografts Models 116
7.7	Examples of Recently Developed Orthotopic Models of Human Cancers 119
7.8	Factors that Can Affect the Usefulness of Preclinical Models in Evaluating New Therapies 120
7.9	Monitoring Metastatic Disease Progression in Preclinical Models 120
7.10	Alternative Preclinical Models: PDX and GEMMs 121
7.11	Recommendations for the Evaluation of Anti-cancer Drugs Using Preclinical Models 122
7.12	Summary 123 References 124
8	Spontaneous Animal Tumor Models 129 <i>Andreas Pospischil, Katrin Grüntzig, Ramona Graf, and Gianluca Boo</i>
8.1	Introduction 129
8.2	Advantages of Spontaneous Canine/Feline Cancer Registries 130
8.2.1	Effective and Relevant Canine/Feline Cancer Registries – Necessary Steps and Existing Registries 131
8.2.1.1	Regional/National/International Population-based Human Cancer Registry with Sufficient Case Numbers and Patient Data 131
8.2.1.2	Regional/National Population-based Canine/Feline Cancer Registries 132
8.2.1.3	Comparative (Human/Canine/Feline) Geographic and Environmental Risk Assessment of Tumor Incidences 133
8.2.1.4	Tissue/Bio-bank Containing Canine/Feline Tumor Samples (Fresh Frozen, FFPE) for Necessary Re-Evaluation, and Further Testing 133
8.2.1.5	Comparative Testing of Genetic/Proteomic Tumor Markers on Different Tumor Tissue from Human and Animal Patients 134
8.3	Spontaneous Animal Tumors as Suitable Models for Human Cancers 134
8.3.1	Canine Tumors 134
8.3.2	Feline Tumors 134
8.4	The Swiss Canine/Feline Cancer Registry 1955–2008 135
8.4.1	Swiss Canine Cancer Registry 1955–2008 135
8.4.1.1	Tumor Location 135
8.4.1.2	Malignancy of the Most Common Tumor Diagnoses 136
8.4.1.3	Sex Distribution 136

8.4.1.4	Breed Distribution	138
8.4.1.5	Sample Catchment Area	140
8.4.2	The Swiss Feline Cancer Registry 1964–2008	140
8.4.2.1	Malignancy of the Most Common Tumor Diagnoses	141
8.4.2.2	Breed Distribution	141
8.4.2.3	Sex Distribution	142
8.4.2.4	Most Common Locations of Tumors (1%)	144
8.4.2.5	Catchment Area	144
8.4.3	Comparison of Swiss Canine, Feline, and Human Cancer Registry Data	146
8.4.4	Conclusion	147
	References	148
9	Dog Models of Naturally Occurring Cancer	153
	<i>Joelle M. Fenger, Jennie Lynn Rowell, Isain Zapata, William C. Kisseberth, Cheryl A. London, and Carlos E. Alvarez</i>	
9.1	Introduction	153
9.1.1	Animal Models of Human Disease and the Need for Alternatives to the Mouse	153
9.2	Advantages of Spontaneous Cancer Models in Dogs	155
9.2.1	High Level of Evolutionary Conservation with Humans	156
9.2.2	Reduced Heterogeneity within Breeds and Increased Variation across Breeds	157
9.2.3	Potential for Comprehensive Genotyping	163
9.2.4	Understanding Both Somatic and Germline Cancer Genetics	164
9.2.5	Translational Models	169
9.3	Dog Cancer Models	170
9.3.1	Canine Cancer Incidence	170
9.3.2	Genetics of Breed-Specific Cancer Models	177
9.3.2.1	Lymphoma	177
9.3.2.2	Osteosarcoma	181
9.4	Preclinical and Veterinary Translational Investigations in Dogs with Cancer	184
9.4.1	Preclinical Investigations in Dogs with Spontaneous Cancer	184
9.4.2	Conduct of Preclinical and Translational Studies in Pet Dogs with Cancer	186
9.4.3	Examples of Successful Preclinical Investigations in Pet Dogs with Cancer	190
9.5	Necessary Developments for Realizing the Potential of Canine Models	196
9.5.1	Epidemiology, Longitudinal Cohorts, Tissue Repositories, and Integrative Genomics	196
9.5.2	Improved Genome Annotation and Development of Key Research Areas	196

9.5.3	Opportunities for Understanding the Complete Biology of Spontaneous Cancers	197
9.5.4	Development of High-Impact Programs in Preclinical Cancer Studies	198
9.6	Key Challenges and Recommendations for Using Canine Models	200
9.6.1	Challenges of Population Structure in Dog Models	200
9.6.2	Recommendations for Optimal Results in Canine Preclinical Research	201
9.7	Conclusions	202
	References	203
10	Improving Preclinical Cancer Models: Lessons from Human and Canine Clinical Trials of Metronomic Chemotherapy	223
	<i>Guido Bocci, Esther K. Lee, Anthony J. Mutsaers, and Urban Emmenegger</i>	
10.1	Introduction: Low-dose Metronomic Chemotherapy	223
10.2	Clinical Trials of Metronomic Chemotherapy	224
10.2.1	Achievements	224
10.2.2	Challenges	225
10.3	Veterinary Metronomic Trials in Pet Dogs with Cancer	227
10.3.1	Adjuvant Treatment	228
10.3.2	First-Line Therapy for Metastatic Disease	229
10.3.3	Biomarker Studies	229
10.3.4	Other Chemotherapy Drug Choices	230
10.3.5	Combination with Targeted Anti-angiogenic Drugs	230
10.3.6	Combining Metronomic and MTD Protocols	231
10.4	Lessons Learned from Clinical Trials: Improving the Predictability of Preclinical Models	231
10.4.1	Pharmacokinetic and Pharmacodynamic Studies in Preclinical Models	231
10.4.1.1	Pharmacokinetic Preclinical Studies of Metronomic Chemotherapy Regimens	233
10.4.1.2	Pharmacodynamic Analyses in Preclinical Studies	236
10.4.2	Pharmacogenomics in Animal Models	237
10.4.3	Pharmacoeconomics of Metronomic Chemotherapy	238
10.5	Conclusions	240
	Acknowledgements	240
	References	240
	Index	247