

# Contents

<i>Preface</i> .....	v
<i>Chapter 1. Adeno-associated viral vectors for gene therapy</i> .....	1
T. R. Flotte and K. I. Berns	
1.1. Biological properties of adeno-associated virus .....	1
1.2. AAV-based gene therapy vectors .....	2
1.3. In vivo applications of rAAV .....	4
1.4. Clinical experience with rAAV .....	7
1.5. Persistence of rAAV vectors .....	8
1.6. Safety of rAAV vector delivery .....	11
1.7. Host range, alternate serotypes, and capsid modifications .....	12
1.8. Remaining questions .....	13
References .....	13
<i>Chapter 2. Production of research and clinical-grade recombinant adeno-associated virus vectors</i> .....	19
J. D. Francis and R. O. Snyder	
2.1. Adeno-associated virus biology .....	20
2.2. rAAV preparation .....	24
2.3. Protocols .....	27
2.4. Small-scale rAAV 1, 2, and 5 vector purification .....	28
2.5. Large-scale rAAV purification .....	30
2.6. rAAV vector characterization .....	31
2.7. Safety testing .....	34
2.8. Pre-clinical regulatory compliance activities .....	35
2.9. Manufacture of clinical-grade rAAV vectors .....	38
2.10. Clinical manufacturing regulatory compliance activities .....	44
References .....	49

<i>Chapter 3. Gene therapy for hemophilia.</i>	57
C. Mah	
3.1. Non-viral DNA vectors . . . . .	59
3.2. Adenovirus vectors . . . . .	60
3.3. Retrovirus vectors . . . . .	62
3.4. Adeno-associated virus vectors . . . . .	64
3.5. Immunological considerations . . . . .	69
3.6. Laboratory protocols . . . . .	72
References . . . . .	73
<i>Chapter 4. Recombinant AAV vectors for gene transfer to the lung: a compartmental approach.</i>	83
T. R. Flotte	
4.1. Introduction . . . . .	83
4.2. Genes, targets and vectors for the lung . . . . .	84
4.3. Therapies targeting the alveoli . . . . .	85
4.4. Therapies targeting the airways . . . . .	87
4.5. Therapies targeting the pulmonary vasculature and pleura . . . . .	92
4.6. Future directions . . . . .	93
References . . . . .	94
<i>Chapter 5. Adeno-associated virus mediated gene therapy for vascular retinopathies.</i>	103
B. J. Raisler, W.-T. Deng, K. I. Berns and W. W. Hauswirth	
5.1. Introduction . . . . .	103
5.2. New strategies for treating NV . . . . .	106
5.3. Protocols . . . . .	114
5.4. Discussion . . . . .	118
References . . . . .	119

<i>Chapter 6. Gene therapy for prevention and treatment of type 1 diabetes.....</i>	125
M. H. Kapturczak, B. R. Burkhardt and M. A. Atkinson	
6.1. The clinical problem diabetes .....	125
6.2. Transplantation .....	126
6.3. Allograft rejection: mechanisms for increasing graft acceptance .....	127
6.4. Recurrent autoimmunity as a mechanism of $\beta$ cell allograft failure .....	129
6.5. Gene transfer into islet cells .....	131
6.6. Potential utility of rAAV-mediated gene therapy for islet transplantation and prevention of autoimmunity recurrence in type 1 diabetes .....	136
6.7. Progress in insulin replacement strategies utilizing gene therapy .....	142
6.8. Summary and future directions .....	146
References .....	147
<i>Chapter 7. Gene therapy for kidney diseases.....</i>	161
S. Chen, K. M. Madsen, C. C. Tisher and A. Agarwal	
7.1. Structure-function correlations .....	162
7.2. Vector systems for gene delivery .....	163
7.3. Methods of gene delivery .....	170
7.4. Targeting specific cells in the kidney .....	172
7.5. Application of gene therapy for specific kidney diseases.....	176
References .....	185
<i>Chapter 8. AAV for disorders of the CNS .....</i>	193
C. Burger, R. J. Mandel and N. Muzyczka	
8.1. Introduction .....	193
8.2. Parkinson disease (PD).....	198
8.3. Alzheimer's disease (AD) .....	204
8.4. Epilepsy .....	206

8.5.	Lysosomal storage disorders (LSD) . . . . .	208
8.6.	Conclusion . . . . .	212
	References . . . . .	213
<i>Chapter 9. Gene therapy for cardiovascular applications . . . . .</i>		225
C. A. Pacak, C. Mah and B. J. Byrne		
9.1.	Viral gene delivery systems . . . . .	227
9.2.	Non-viral gene delivery systems . . . . .	232
9.3.	Gene delivery route . . . . .	233
9.4.	Cellular and gene therapy combinations . . . . .	234
9.5.	Conclusions . . . . .	234
9.6.	Methods . . . . .	235
	References . . . . .	239
<i>Chapter 10. Gene therapy for lysosomal storage disorders . . . . .</i>		243
K. O. Cresawn and B. J. Byrne		
10.1.	The lysosome . . . . .	243
10.2.	Lysosomal storage diseases . . . . .	244
10.3.	Current therapies . . . . .	244
10.4.	Gene therapy . . . . .	245
10.5.	Glycogen storage disease type II . . . . .	248
10.6.	Gene therapy for GSD II: Proof of concept studies . . . . .	254
10.7.	Recombinant adeno-associated virus vector studies . . . . .	256
10.8.	Recombinant AAV-mediated treatment of GSDII . . . . .	257
10.9.	Gene therapy for CNS pathologies in LSDs . . . . .	262
10.10.	Conclusion . . . . .	265
	References . . . . .	266
<i>Index . . . . .</i>		277