

Cambridge University Press

0521425034 - Monoclonal Antibodies: Production, Engineering and Clinical Application

Edited by Mary A. Ritter and Heather M. Ladyman

Frontmatter

[More information](#)

Monoclonal antibodies have an important and wide-ranging role in many areas of biomedical research and this volume is among the first to combine technical and clinical aspects of the subject. Monoclonal antibodies provide highly specific reagents with which to identify, analyse, quantitate and manipulate molecules, both in solution and in solid phase, such as at a cell surface. The aim of this book is to provide a unique combination of information concerning the production (by both cellular and molecular biology techniques), and structural and functional characteristics of monoclonal antibodies, together with detailed discussions of the various analytic, diagnostic and therapeutic applications of these antibodies in many areas of clinical medicine, including histopathology, oncology, transplantation, infectious diseases, rheumatology, haematology and dermatology.

Cambridge University Press

0521425034 - Monoclonal Antibodies: Production, Engineering and Clinical Application

Edited by Mary A. Ritter and Heather M. Ladyman

Frontmatter

[More information](#)

Monoclonal antibodies

Cambridge University Press

0521425034 - Monoclonal Antibodies: Production, Engineering and Clinical Application

Edited by Mary A. Ritter and Heather M. Ladyman

Frontmatter

[More information](#)

POSTGRADUATE MEDICAL SCIENCE

This important new series is based on the successful and internationally well-regarded specialist training programme at the Royal Postgraduate Medical School in London. Each volume provides an integrated and self-contained account of a key area of medical science, developed in conjunction with the course organisers and including contributions from specially invited authorities.

The aim of the series is to provide biomedical and clinical scientists with a reliable introduction to the theory and to the technical and clinical applications of each topic.

The volumes will be a valuable resource and guide for trainees in the medical and biomedical sciences and for laboratory-based scientists.

In the series:

Radiation protection of patients by R. Wootton

Image analysis in histology: conventional and confocal microscopy by
D. Springall, R. Wootton and J. Polak

Cambridge University Press

0521425034 - Monoclonal Antibodies: Production, Engineering and Clinical Application

Edited by Mary A. Ritter and Heather M. Ladyman

Frontmatter

[More information](#)

POSTGRADUATE MEDICAL SCIENCE

Monoclonal antibodies

Production, engineering and clinical application

EDITED BY

MARY A. RITTER and
HEATHER M. LADYMAN

*Department of Immunology
Royal Postgraduate Medical School*

Published in association with the Royal Postgraduate Medical School
University of London by



Cambridge University Press

0521425034 - Monoclonal Antibodies: Production, Engineering and Clinical Application

Edited by Mary A. Ritter and Heather M. Ladyman

Frontmatter

[More information](#)

CAMBRIDGE UNIVERSITY PRESS

Cambridge, New York, Melbourne, Madrid, Cape Town, Singapore, São Paulo

Cambridge University Press

The Edinburgh Building, Cambridge CB2 2RU, UK

Published in the United States of America by Cambridge University Press, New York

www.cambridge.org

Information on this title: www.cambridge.org/9780521473545

© Cambridge University Press 1995

This publication is in copyright. Subject to statutory exception and to the provisions of relevant collective licensing agreements, no reproduction of any part may take place without the written permission of Cambridge University Press.

First published 1995

A catalogue record for this publication is available from the British Library

ISBN-13 978-0-521-47354-5 hardback

ISBN-10 0-521-47354-3 hardback

ISBN-13 978-0-521-42503-2 paperback

ISBN-10 0-521-42503-4 paperback

Transferred to digital printing 2005

Contents

1	Introduction	1
	<i>M. A. Ritter</i>	
1.1	Introduction	1
1.2	Generation of an immune response	1
1.3	Antibody structure	2
1.4	Generation of the B cell repertoire	4
1.5	What is a monoclonal antibody?	6
1.6	The aim of this book	8
1.7	References	8
2	Production of monoclonal antibodies	9
	<i>H. M. Ladyman and M. A. Ritter</i>	
2.1	Introduction	9
2.2	Antigens and immunization	9
2.3	Cell fusion	15
2.4	Growth and selection of hybrids	18
2.5	Cloning techniques	22
2.6	Cell maintenance and bulk production of antibody	24
2.7	Characterization and purification of antibody	26
2.8	Storage	30
2.9	Analysis of the structure of the antigen to which your monoclonal antibody binds	31
2.10	Summary	32
2.11	References	32
3	Antigen–antibody interactions: how affinity and kinetics affect assay design and selection procedures	34
	<i>R. J. Morris</i>	
3.1	Introduction	34
3.2	What is affinity?	34
3.3	Kinetics of association and dissociation	37
3.4	Sensitivity, and its relationship to affinity	41
3.5	Other parameters affecting affinity	44
3.6	The nature of a protein epitope and its influence on antibody binding	50

Cambridge University Press

0521425034 - Monoclonal Antibodies: Production, Engineering and Clinical Application

Edited by Mary A. Ritter and Heather M. Ladyman

Frontmatter

[More information](#)

viii	<i>Contents</i>
3.7	So what is antibody specificity? 53
3.8	Summary 56
3.9	References 57
4	Production and characterization of synthetic peptide-derived antibodies 60
	<i>K. M. Price</i>
4.1	Introduction 60
4.2	Prediction and selection of antigenic determinants 61
4.3	Conjugation 63
4.4	Immunization 66
4.5	Screening 66
4.6	Epitope detection and analysis 69
4.7	Appendix 78
4.8	References 82
5	Human monoclonal antibodies: production, use, problems 85
	<i>A. Sa'adu and A. Zumla</i>
5.1	Production of human monoclonal antibodies 85
5.2	Use of human monoclonal antibodies 90
5.3	Problems with human monoclonal antibody production 97
5.4	Conclusion 106
5.5	References 106
6	Bispecific monoclonal antibodies 121
	<i>S. Songvilai and P. J. Lachmann</i>
6.1	Introduction 121
6.2	Production of bispecific monoclonal antibodies 123
6.3	Applications of bispecific antibodies: current status and future development 132
6.4	References 136
7	Production of antibodies using phage display libraries 142
	<i>A. J. T. George</i>
7.1	Introduction 142
7.2	Filamentous phage 143
7.3	Filamentous phage as expression vectors 144
7.4	Antibody selection on phage 146
7.5	Recombinant antibody fragments 154
7.6	Conclusions 159
7.7	References 160
8	Genetic manipulation of monoclonal antibodies 166
	<i>N. S. Courtenay-Luck</i>
8.1	Introduction 166
8.2	Humanization 166
8.3	Recombinant antibody fragments 170
8.4	Discussion 175
8.5	References 177

<i>Contents</i>		ix
9	Monoclonal antibodies in diagnostic immunoassays <i>D. B. Cook and C. H. Self</i>	180
9.1	Introduction	180
9.2	Advantages and pitfalls of using monoclonal antibodies	183
9.3	Specificity	185
9.4	Small molecular weight analytes	188
9.5	Endogenous interference in immunoassays	190
9.6	High-dose hook phenomenon	193
9.7	Use of established labelling technology	194
9.8	Sensitivity	194
9.9	Speed	195
9.10	Convenience	196
9.11	Methods of detection	197
9.12	Fibre optic sensors	201
9.13	Future of monoclonal antibodies in diagnostics	201
9.14	References	203
10A	Monoclonal antibodies in oncology: diagnostic pathology <i>A. Colfor and P. A. Hall</i>	209
10A.1	Introduction	209
10A.2	Genotype and phenotype	209
10A.3	Use of immunohistochemistry in diagnostic pathology	210
10A.4	Use of immunohistochemistry in diagnosis	210
10A.5	Immunohistochemistry as an educational aid	214
10A.6	Use in prognosis	214
10A.7	Histogenesis versus differentiation	216
10A.8	Future prospects	218
10A.9	Conclusion	219
10A.10	References	219
10B	Monoclonal antibodies in oncology: <i>in-vivo</i> targeting for immunoscintigraphy and therapy of human malignancies <i>A. Bamias and A. A. Epenetos</i>	222
10B.1	Introduction	222
10B.2	Immunoscintigraphy	223
10B.3	Immunotherapy	229
10B.4	Conclusion	238
10B.5	References	238
11A	Monoclonal antibodies in infectious disease: diagnosis <i>T. G. Wreghitt and J. J. Gray</i>	247
11A.1	Introduction	247
11A.2	Respiratory infections	247
11A.3	Gastroenteritis	248
11A.4	Hepatitis	249
11A.5	Neurological/congenital infections	250
11A.6	Meningitis	250
11A.7	Encephalitis	251
11A.8	Sexually transmitted disease	252

x	<i>Contents</i>
11A.9	Infection in the immunocompromised host 253
11A.10	References 255
11B	Monoclonal antibodies in infectious disease: prophylaxis and therapy 260
	<i>J. Cohen</i>
11B.1	Introduction 260
11B.2	Bacterial infections 261
11B.3	Viral and chlamydial infections 266
11B.4	Additional applications of monoclonal antibodies 268
11B.5	Conclusions 268
11B.6	References 269
12A	Monoclonal antibodies in transplantation: immunohistology 274
	<i>M. L. Rose</i>
12A.1	Introduction 274
12A.2	Nature of the infiltrate 274
12A.3	Major histocompatibility complex antigens 276
12A.4	Adhesion molecules 285
12A.5	Cytokines 287
12A.6	References 288
12B	Experimental studies on <i>in-vivo</i> immunosuppression 292
	<i>N. M. Parish and A. Cooke</i>
12B.1	Introduction 292
12B.2	Monoclonal antibody treatments directed towards T cell surface markers 294
12B.3	Monoclonal antibody therapies targeted at antigen-presenting cells 304
12B.4	Monoclonal antibody therapies targeting cytokines 305
12B.5	Monoclonal antibody therapies targeting adhesion molecules 308
12B.6	Monoclonal antibodies directed towards miscellaneous cell markers 309
12B.7	References 310
12C	Monoclonal antibodies in transplantation: prophylaxis and treatment of graft-versus-host disease after bone marrow transplantation 317
	<i>L. Boström and O. Ringden</i>
12C.1	Introduction 317
12C.2	Prevention of GVHD with <i>in-vitro</i> use of monoclonal antibodies 319
12C.3	Randomized clinical studies comparing pharmacological GVHD prophylaxis with T cell depletion 324
12C.4	Monoclonal antibodies <i>in vivo</i> for prevention of GVHD 325
12C.5	Treatment of GVHD with monoclonal antibodies 326
12C.6	Monoclonal antibodies in the diagnosis of GVHD 328
12C.7	Outlook 329
12C.8	References 329

Cambridge University Press

0521425034 - Monoclonal Antibodies: Production, Engineering and Clinical Application

Edited by Mary A. Ritter and Heather M. Ladyman

Frontmatter

[More information](#)

<i>Contents</i>		xi
12D	Monoclonal antibodies in transplantation: use as therapeutic agents in clinical organ transplantation <i>M. Giral, J. Dantal, D. Cantarovich, M. Hourmant, R. Baatard, B. Le Mauff, Y. Jacques and J. P. Souillou</i>	340
12D.1	Introduction	340
12D.2	Monoclonal antibodies reacting with all T lymphocytes	341
12D.3	Antibodies that react against a subset of T lymphocytes	346
12D.4	Monoclonal antibodies against interleukin-2 receptors	347
12D.5	Engineered monoclonal antibody molecules: chimeric, 'humanized' and fusion proteins	351
12D.6	Comments	352
12D.7	Acknowledgements	353
12D.8	References	353
13	Monoclonal antibodies and the skin <i>A. C. Chu and E. Tsele</i>	362
13.1	Introduction	362
13.2	Structure of the skin	362
13.3	The keratinocyte	362
13.4	Melanocytes	366
13.5	Langerhans cells	367
13.6	Basement membrane zone	368
13.7	Dermal infiltrates	370
13.8	Cutaneous T cell lymphoma	371
13.9	Benign cellular infiltrates	371
13.10	Adhesion molecules	372
13.11	References	373
14	Monoclonal antibodies in endocrinology <i>E. Hillhouse and C. H. Self</i>	380
14.1	Introduction	380
14.2	Assays of hormones	380
14.3	Generation of suitable monoclonal antibodies	382
14.4	Use of monoclonal antibodies	383
14.5	Specific modification	393
14.6	References	396
15	Monoclonal antibodies in rheumatology <i>J. D. Isaacs</i>	403
15.1	Introduction	403
15.2	Normal immune response	403
15.3	Monoclonal antibodies to control immune responsiveness	406
15.4	Animal models of rheumatological disorders	408
15.5	Monoclonal antibodies in human rheumatic diseases	410
15.6	Adverse effects of monoclonal antibodies	417
15.7	The future	419
15.8	Summary	420
15.9	Acknowledgements	421
15.10	References	421

Cambridge University Press

0521425034 - Monoclonal Antibodies: Production, Engineering and Clinical Application

Edited by Mary A. Ritter and Heather M. Ladyman

Frontmatter

[More information](#)

xii	<i>Contents</i>
16	Technical appendix: monoclonal antibody production methods 429
	<i>H. M. Ladyman and M. A. Ritter</i>
16.1	Immunization 429
16.2	Media and general cell culture 429
16.3	Contamination in tissue culture 430
16.4	Cryopreservation of cells 431
16.5	The fusion 433
16.6	Cloning techniques 436
16.7	Bulk production of monoclonal antibodies 440
16.8	Screening techniques 441
16.9	Monoclonal antibody characterization and purification 449
16.10	SDS-PAGE and Western blotting 459
Index	467

Contributors

Dr R. Baatard

Service de Néphrologie, Immunologie Clinique et Unité de Recherche,
INSERM U. 211, CHU Nantes, France

Dr A. Bamias

Imperial Cancer Research Fund Laboratories, Hammersmith Hospital, Du
Cane Road, London W12 0NN, UK

Dr L. Boström

Department of Clinical Immunology, The Karolinska Institute, Huddinge
Hospital, Stockholm, Sweden

Dr D. Cantarovich

Service de Néphrologie, Immunologie Clinique et Unité de Recherche,
INSERM U. 211, CHU Nantes, France

Dr A. C. Chu

Unit of Dermatology, Hammersmith Hospital, Du Cane Road, London W12
0NN, UK

Dr J. Cohen

Department of Bacteriology, Royal Postgraduate Medical School, Du Cane
Road, London W12 0NN, UK

Dr A. Colfor

Department of Histopathology, Royal Marsden Hospital, Fulham Road,
London SW3 6JJ, UK

Dr D. B. Cook

Department of Clinical Biochemistry, The Medical School, Framlingham
Place, Newcastle-upon-Tyne, NE2 4HH, UK

Dr A. Cooke

Immunology Division, Department of Pathology, Cambridge University,
Tennis Court Road, Cambridge CB2 1QP, UK

Dr N. S. Courtenay-Luck

Antisoma plc, Samaritan Hospital for Women, 153–173 Marylebone Road,
London NW1 5QH, UK

Cambridge University Press

0521425034 - Monoclonal Antibodies: Production, Engineering and Clinical Application

Edited by Mary A. Ritter and Heather M. Ladyman

Frontmatter

[More information](#)

xiv

Contributors

Dr J. Dantal

Service de Néphrologie, Immunologie Clinique et Unité de Recherche,
INSERM U. 211, CHU Nantes, France

Dr A. A. Epenetos

Imperial Cancer Research Fund Laboratories, MRC Building, Hammersmith
Hospital, Du Cane Road, London W12 0HS, UK

Dr A. J. T. George

Department of Immunology, Royal Postgraduate Medical School, Du Cane
Road, London W12 0NN, UK

Dr M. Giral

Service de Néphrologie, Immunologie Clinique et Unité de Recherche,
INSERM U. 211, CHU Nantes, France

Dr J. J. Gray

Clinical Microbiology and Public Health Laboratory, Level 6, Addenbrookes
Hospital, Hills Road, Cambridge CB2 2QW, UK

Prof. P. A. Hall

Pathology Department, Ninewells Hospital and Medical School, University of
Dundee, Dundee DD1 9SY, UK

Dr E. Hillhouse

Department of Clinical Biochemistry and Metabolic Medicine, The Medical
School, Framlingham Place, Newcastle-upon-Tyne NE2 4HH, UK

Dr M. Hourmant

Service de Néphrologie, Immunologie Clinique et Unité de Recherche,
INSERM U. 211, CHU Nantes, France

Dr J. D. Isaacs

Division of Immunology, Department of Pathology, Cambridge University,
Tennis Court Road, Cambridge CB2 1QP, UK

Dr Y. Jacques

Service de Néphrologie, Immunologie Clinique et Unité de Recherche,
INSERM U. 211, CHU Nantes, France

Prof. P. J. Lachmann

Molecular Immunopathology Unit, Medical Research Council Centre and
University of Cambridge Clinical School, Cambridge CB2 2QH, UK

Ms H. M. Ladyman

Department of Immunology, Royal Postgraduate Medical School, Du Cane
Road, London W12 0NN, UK

Dr B. Le Mauff

Service de Néphrologie, Immunologie Clinique et Unité de Recherche,
INSERM U. 211, CHU Nantes, France

Dr R. J. Morris

Laboratory of Neurobiology, National Institute for Medical Research, The
Ridgeway, Mill Hill, London NW7 1AA, UK

Cambridge University Press

0521425034 - Monoclonal Antibodies: Production, Engineering and Clinical Application

Edited by Mary A. Ritter and Heather M. Ladyman

Frontmatter

[More information](#)

Contributors

xv

Dr N. M. Parish

Division of Immunology, Department of Pathology, University of Cambridge,
Tennis Court Road, Cambridge CB2 1QP, UK

Mr K. M. Price

Zeneca, Cambridge Research Biochemicals Ltd, Gadbrook Park, Northwich,
Cheshire CW9 7RA, UK

Prof. O. Ringden

Department of Clinical Immunology, The Karolinska Institute, Huddinge
Hospital, Stockholm, Sweden

Prof. M. A. Ritter

Department of Immunology, Royal Postgraduate Medical School, Du Cane
Road, London W12 0NN, UK

Dr M. L. Rose

Immunology Department, Harefield Hospital, Harefield, Uxbridge UB9 6JH,
UK

Dr A. Sa'adu

Division of Immunological Medicine, Clinical Research Centre, Northwick
Park Hospital, Watford Road, Harrow HA1 3UJ, UK

Prof. C. H. Self

Department of Clinical Biochemistry and Metabolic Medicine, The Medical
School, Framlingham Place, Newcastle-upon-Tyne, NE2 4HH, UK

Dr S. Songsivilai

Department of Immunology, Faculty of Medicine, Siriraj Hospital, Mahidol
University, Bangkok 10700, Thailand

Dr J. P. Souillou

Service de Néphrologie, Immunologie Clinique et Unité de Recherche,
INSERM U. 211, CHU Nantes, France

Dr E. Tsele

Department of Dermatology, Royal Postgraduate Medical School, Du Cane
Road, London W12 0NN, UK

Dr T. G. Wreghitt

Clinical Microbiology and Public Health Laboratory, Level 6,
Addenbrookes Hospital, Hills Road, Cambridge CB2 2QW, UK

Dr A. Zumla

School of Medicine, University Teaching Hospital, PO Box 50110, Lusaka,
Zambia