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Antibody engineering

A Practical Approach

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IRL PRESS
at
OXFORD UNIVERSITY PRESS
Oxford New York Tokyo

Oxford University Press, Walton Street, Oxford OX2 6DP

Oxford New York
Athens Auckland Bangkok Bombay
Calcutta Cape Town Dar es Salaam Delhi
Florence Hong Kong Istanbul Karachi
Kuala Lumpur Madras Madrid Melbourne
Mexico City Nairobi Paris Singapore
Taipei Tokyo Toronto
and associated companies in
Berlin Ibadan

Oxford is a trade mark of Oxford University Press

Published in the United States
by Oxford University Press Inc., New York

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

Library of Congress Cataloging in Publication Data
Antibody engineering: a practical approach/edited by John McCafferty, Hennie Hogenboom, and Dave Chiswell. – 1st ed.
(A Practical approach series ; 169)
Includes bibliographical references and index.
1. Immunoglobulins–Synthesis–Laboratory manuals. 2. Genetic engineering–Laboratory manuals. I. McCafferty, John, Dr. II. Hoogenboom, Hennie III. Chiswell, Dave. IV. Series.
QR186.7.A578 1996 615'.37–dc20 95–49218

ISBN 0 19 963593 5 (Hbk)
ISBN 0 19 963592 7 (Pbk)

Typeset by Footnote Graphics, Warminster, Wilts
Printed in Great Britain by Information Press, Ltd, Eynsham, Oxon.

Preface

Recombinant DNA methods have revolutionized the isolation and production of antibodies in recent years. Initially, recombinant antibodies were engineered and expressed in mammalian cells but by the late 1980s efficient expression of antibody fragments in bacteria had been demonstrated. More recently, the isolation of new antibody specificities has been transformed by the creation of large repertoires of antibody genes and the development of methods based on bacteriophage display to isolate specific antibodies. This has been paralleled by an increased understanding of the key residues affecting antibody structure and specificity together with more quantitative analysis of the binding properties of the antibodies. In addition, the mapping and sequencing of the repertoire of human antibody germline genes has put our understanding of the changes occurring during *in vivo* affinity maturation on a firmer footing.

We believe that now is the right time to assemble in one volume, protocols which allow the researcher to isolate a new antibody, analyse its properties, format the right antibody molecule or fragment, and produce sufficient quantities to be useful.

The book is structured in two parts: the first describes the generation and analysis of antibodies and the second covers engineering and production. Chapter 1 is designed to be the source of the standard 'repertoire' of phage-display protocols. In the following chapter the 'fine art' of the system, e.g. affinity maturation, is described. Methods for introducing antibody repertoires into transgenic mice are also described in the first section, as are methods for the analysis of the characteristics of antibodies with respect to affinity and sequence.

The second section describes the engineering of natural and man-made effector functions. The conversion of rodent antibodies to human antibodies by either CDR grafting or 'guided selection' using phage display is also covered. In addition, methods to manufacture significant quantities of various antibody-based molecules in both eukaryotic and prokaryotic systems are outlined.

Traditional monoclonal antibody technology has provided a wealth of reagents for research and diagnostic applications. The methods and technologies described here can help expand the versatility of antibody-based reagents in these areas. We have endeavoured in this book to provide researchers working with antibodies in research and diagnostics with a route into the 'new technologies', as practised by the leaders in the field.

This 'route' will be of equal value to workers in the field of antibody therapeutics. The ability to generate high-affinity, high-specificity human antibodies in appropriate formats will help circumvent some of the problems

associated with earlier work using mouse monoclonal antibodies and aid in their successful use *in vivo*. As we draw to the end of the millennium we have moved closer to the 'magic bullet' first hypothesized by Paul Ehrlich at the beginning of this century.

Cambridge J.McC.
Maastricht H.R.H.
May 1996 D.J.C.

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