

ADVANCES IN

# Immunology

Vol. 13

*EDITED BY*

F. J. DIXON, JR.

HENRY G. KUNKEL

ADVANCES IN

# Immunology

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## Erratum

### ADVANCES IN IMMUNOLOGY

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F. J. Dixon and Henry G. Kunkel

On page 28, the following line should be inserted between lines 12 and 13:  
"P-K, PCA, and passive sensitization of leukocytes, which, therefore"

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## PREFACE

It can safely be predicted that the seventies will be the decade of cellular immunology. Already the expansion is evident on all sides and many immunologists, previously involved in the antibody field, are turning to cellular work. To some extent this may be unfortunate because that most basic of problems, the mechanism of antibody variability, remains an enigma. Immunologists are still evenly divided on the issue of whether the germ line theory or a type of somatic model holds the explanation. Possibly the work at the cellular level might provide the answer. The *Advances* will participate actively in this timely movement and, as exemplified by this volume, will also continue to be involved in the currently less popular but no less important areas of immunology.

The article by Drs. John E. Hopper and Alfred Nisonoff concerns that very useful label of the immunoglobulins, their individual antigenic specificity or idiotypic specificity. The authors have utilized this property in superb fashion to trace the development of different antibody producing clones of cells in the primary and secondary response. It is abundantly clear that antigens related to the V regions and antibody combining sites are followed in these studies.

There are few areas where problems of nomenclature are more varied and confusing than in the field of allergic reactions. Dr. Elmer L. Becker treats this subject from all aspects, ranging from the initiating antigen, through the mediators produced, to the final cell involvement. A very reasonable classification of immediate-type allergic reactions has emerged that takes into account the many different phases of these reactions.

One of the exciting chapters in immunology has been the recent delineation of the IgE class of immunoglobulins and the demonstration of its significance for atopic allergic disorders. Just as in all other areas of immunoglobulin work, the discovery of a myeloma protein of the IgE type contributed enormously to the successful evolution of this work. Drs. Hans Bennich and S. Gunnar O. Johansson were responsible for this important aspect and they not only review this field but also present many observations that have not been published elsewhere. Because of the low concentration of IgE in most sera, its measurement has presented a special challenge. The ingenious procedures developed by the authors, as well as other methods, are discussed in useful detail.

Drs. J. L. Turk and A. D. M. Bryceson review the various different immunological reactions to the specific organisms in leprosy and

leishmaniasis. These authors have played a primary role in interpreting these reactions in terms of modern concepts of immunology. Defects in cellular immunity clearly play a major role in special forms of these disorders and many of the principles derived from these studies hold implications for a number of other diseases.

Dr. Barry R. Bloom, one of the leaders in the cellular immunity expansion, describes some of the forefronts of this field. The many mediators involved in lymphocyte reactions are considered in special detail. None of these factors has been isolated in pure form, which will be essential for their eventual understanding. However, an overall picture of the intricacies of cellular immunity is beginning to emerge which relates the various experimental models to *in vivo* events.

The cooperation and valuable assistance of the publishers in the production of Volume 13 are gratefully acknowledged.

H. G. KUNKEL

F. J. DIXON

July 1971

## Contents of Previous Volumes

### Volume 1

Transplantation Immunity and Tolerance

M. HAŠEK, A. LENGEROVÁ, AND T. HRABA

Immunological Tolerance of Nonliving Antigens

RICHARD T. SMITH

Functions of the Complement System

ABRAHAM G. OSLER

*In Vitro* Studies of the Antibody Response

ABRAM B. STAVITSKY

Duration of Immunity in Virus Diseases

J. H. HALE

Fate and Biological Action of Antigen-Antibody Complexes

WILLIAM O. WEIGLE

Delayed Hypersensitivity to Simple Protein Antigens

P. G. H. GELL AND B. BENACERRAF

The Antigenic Structure of Tumors

P. A. GORER

AUTHOR INDEX-SUBJECT INDEX

### Volume 2

Immunologic Specificity and Molecular Structure

FRED KARUSH

Heterogeneity of  $\gamma$ -Globulins

JOHN L. FAHEY

The Immunological Significance of the Thymus

J. F. A. P. MILLER, A. H. E. MARSHALL, AND R. G. WHITE

Cellular Genetics of Immune Responses

G. J. V. NOSSAL

Antibody Production by Transferred Cells

CHARLES G. COCHRANE AND FRANK J. DIXON

Phagocytosis

DERRICK ROWLEY

**Antigen-Antibody Reactions in Helminth Infections**

E. J. L. SOULSBY

**Embryological Development of Antigens**

REED A. FLICKINGER

**AUTHOR INDEX-SUBJECT INDEX****Volume 3*****In Vitro* Studies of the Mechanism of Anaphylaxis**

K. FRANK AUSTEN AND JOHN H. HUMPHREY

**The Role of Humoral Antibody in the Homograft Reaction**

CHANDLER A. STETSON

**Immune Adherence**

D. S. NELSON

**Reaginic Antibodies**

D. R. STANWORTH

**Nature of Retained Antigen and Its Role in Immune Mechanisms**

DAN H. CAMPBELL AND JUSTINE S. GARVEY

**Blood Groups in Animals Other Than Man**

W. H. STONE AND M. R. IRWIN

**Heterophile Antigens and Their Significance in the Host-Parasite Relationship**

C. R. JENKIN

**AUTHOR INDEX-SUBJECT INDEX****Volume 4****Ontogeny and Phylogeny of Adoptive Immunity**

ROBERT A. GOOD AND BEN W. PAPERMASTER

**Cellular Reactions in Infection**

EMANUEL SUTER AND HANSRUEDY RAMSEIER

**Ultrastructure of Immunologic Processes**

JOSEPH D. FELDMAN

**Cell Wall Antigens of Gram-Positive Bacteria**

MACLYN MCCARTY AND STEPHEN I. MORSE

**Structure and Biological Activity of Immunoglobulins**

SYDNEY COHEN AND RODNEY R. PORTER



**Autoantibodies and Disease**

H. G. KUNKEL AND E. M. TAN

**Effect of Bacteria and Bacterial Products on Antibody Response**

J. MUNOZ

**AUTHOR INDEX-SUBJECT INDEX**

**Volume 5**

**Natural Antibodies and the Immune Response**

STEPHEN V. BOYDEN

**Immunological Studies with Synthetic Polypeptides**

MICHAEL SELA

**Experimental Allergic Encephalomyelitis and Autoimmune Disease**

PHILIP Y. PATERSON

**The Immunology of Insulin**

C. G. POPE

**Tissue-Specific Antigens**

D. C. DUMONDE

**AUTHOR INDEX-SUBJECT INDEX**

**Volume 6**

**Experimental Glomerulonephritis: Immunological Events  
and Pathogenetic Mechanisms**

EMIL R. UNANUE AND FRANK J. DIXON

**Chemical Suppression of Adaptive Immunity**

ANN E. GABRIELSON AND ROBERT A. GOOD

**Nucleic Acids as Antigens**

OTTO J. PLESCIA AND WERNER BRAUN

***In Vitro* Studies of Immunological Responses of Lymphoid Cells**

RICHARD W. DUTTON

**Developmental Aspects of Immunity**

JAROSLAV ŠTERZL AND ARTHUR M. SILVERSTEIN

**Anti-antibodies**

PHILIP G. H. GELL AND ANDREW S. KELUS

**Conglutinin and Immunoconglutinins**

P. J. LACHMANN

**AUTHOR INDEX-SUBJECT INDEX**

**Volume 7**

Structure and Biological Properties of Immunoglobulins  
SYDNEY COHEN AND CESAR MILSTEIN

Genetics of Immunoglobulins in the Mouse  
MICHAEL POTTER AND ROSE LIEBERMAN

Mimetic Relationships between Group A Streptococci  
and Mammalian Tissues  
JOHN B. ZABRISKIE

Lymphocytes and Transplantation Immunity  
DARCY B. WILSON AND R. E. BILLINGHAM

Human Tissue Transplantation  
JOHN P. MERRILL

AUTHOR INDEX-SUBJECT INDEX

**Volume 8**

Chemistry and Reaction Mechanisms of Complement  
HANS J. MÜLLER-EBERHARD

Regulatory Effect of Antibody on the Immune Response  
JONATHAN W. UHR AND GÖRAN MÖLLER

The Mechanism of Immunological Paralysis  
D. W. DRESSER AND N. A. MITCHISON

*In Vitro* Studies of Human Reaginic Allergy  
ABRAHAM G. OSLER, LAWRENCE M. LICHTENSTEIN, AND DAVID A. LEVY

AUTHOR INDEX-SUBJECT INDEX

**Volume 9**

Secretory Immunoglobulins  
THOMAS B. TOMASI, JR., AND JOHN BIENENSTOCK

Immunologic Tissue Injury Mediated by Neutrophilic Leukocytes  
CHARLES G. COCHRANE

The Structure and Function of Monocytes and Macrophages  
ZANVIL A. COHN

The Immunology and Pathology of NZB Mice  
J. B. HOWIE AND B. J. HELYER

AUTHOR INDEX-SUBJECT INDEX

**Volume 10****Cell Selection by Antigen in the Immune Response**

GREGORY W. SISKIND AND BARUJ BENACERRAF

**Phylogeny of Immunoglobulins**

HOWARD M. GREY

**Slow Reacting Substance of Anaphylaxis**

ROBERT P. ORANGE AND K. FRANK AUSTEN

**Some Relationships among Hemostasis, Fibrinolytic Phenomena, Immunity, and the Inflammatory Response**

OSCAR D. RATNOFF

**Antigens of Virus-Induced Tumors**

KARL HABEL

**Genetic and Antigenetic Aspects of Human Histocompatibility Systems**

D. BERNARD AMOS

**AUTHOR INDEX-SUBJECT INDEX****Volume 11****Electron Microscopy of the Immunoglobulins**

N. MICHAEL GREEN

**Genetic Control of Specific Immune Responses**

HUGH O. McDEVITT AND BARUJ BENACERRAF

**The Lesions in Cell Membranes Caused by Complement**

JOHN H. HUMPHREY AND ROBERT R. DOURMASHKIN

**Cytotoxic Effects of Lymphoid Cells *In Vitro***

PETER PERLMANN AND GÖRAN HOLM

**Transfer Factor**

H. S. LAWRENCE

**Immunological Aspects of Malaria Infection**

IVOR N. BROWN

**AUTHOR INDEX-SUBJECT INDEX****Volume 12****The Search for Antibodies with Molecular Uniformity**

RICHARD M. KRAUSE

**Structure and Function of  $\gamma$ M Macroglobulins**

HENRY METZGER

**Transplantation Antigens****R. A. REISFELD AND B. D. KAHAN****The Role of Bone Marrow in the Immune Response****NABIH I. ABDON AND MAXWELL RICHTER****Cell Interaction in Antibody Synthesis****D. W. TALMAGE, J. RADOVICH, AND H. HEMMINGSEN****The Role of Lysosomes in Immune Responses****GERALD WEISSMANN AND PETER DUKOR****Molecular Size and Conformation of Immunoglobulins****KEITH J. DORRINGTON AND CHARLES TANFORD****AUTHOR INDEX-SUBJECT INDEX**

# CONTENTS

LIST OF CONTRIBUTORS . . . . .	vii
PREFACE . . . . .	ix
CONTENTS OF PREVIOUS VOLUMES . . . . .	xi

## Structure and Function of Human Immunoglobulin E

HANS BENNICH AND S. GUNNAR O. JOHANSSON

I. Introduction . . . . .	1
II. Isolation and Physicochemical Characteristics of Immunoglobulin E . . . . .	2
III. Properties of Antigenically and Biologically Active Structural Regions of Immunoglobulin E . . . . .	19
IV. Methods for Determination . . . . .	28
V. Levels of Immunoglobulin E in Healthy Individuals . . . . .	29
VI. Levels of Immunoglobulin E in Disease . . . . .	35
VII. Detection of Antibody Activity in the Immunoglobulin E Class . . . . .	45
VIII. Metabolism . . . . .	49
IX. Concluding Remarks . . . . .	51
References . . . . .	51

## Individual Antigenic Specificity of Immunoglobulins

JOHN E. HOPPER AND ALFRED NISONOFF

I. Introduction . . . . .	58
II. Individual Antigenic Specificities in Monoclonal Proteins . . . . .	60
III. Individual Antigenic Specificities in Antibody Populations . . . . .	63
IV. Cross-Reactions of Antiidiotypic Sera and Evidence for Identical Molecules in Different Individual Animals . . . . .	69
V. Evidence Based on Idiotypic Specificity for Limited Heterogeneity of Normal Antibody Populations . . . . .	75
VI. Persistence and Changes of Antibody Populations during Prolonged Immunization . . . . .	76
VII. Shared Idiotypic Determinants in IgG and IgM Antibodies of the Same Specificity . . . . .	81
VIII. Localization of Individually Specific Antigenic Determinants . . . . .	85
IX. Cross-Reactions of Anti- <i>ind</i> Antibodies with Nonspecific Immunoglobulins . . . . .	92
X. Monoclonal Origin of Molecules with Individually Specific Antigenic Determinants . . . . .	94
XI. Summary . . . . .	95
References . . . . .	97

## *In Vitro* Approaches to the Mechanism of Cell-Mediated Immune Reactions

BARRY R. BLOOM

I. Introduction . . . . .	102
II. Lymphocyte Transformation . . . . .	104

III. Direct. Cytotoxicity of Target Cells by Lymphocytes . . . . .	111
IV. Mediators—Qualitive Basis of the Response . . . . .	122
V. Enumeration of Specifically Sensitized Cells—Quantitative Basis of the Response . . . . .	160
VI. Reality Testing—Relationships between <i>in Vitro</i> Results and Cell-Mediated Immunity <i>in Vivo</i> . . . . .	169
VII. Relationships between Cell-Mediated Immunity and Antibody Formation . . . . .	178
References . . . . .	193

### Immunological Phenomena in Leprosy and Related Diseases

J. L. TURK AND A. D. M. BRYCESON

I. Introduction . . . . .	209
II. Leprosy . . . . .	210
III. Leishmaniasis . . . . .	237
IV. Concept of a Host-Determined Spectrum of Clinical Manifestations in Other Chronic Infections in Man . . . . .	259
References . . . . .	261

### Nature and Classification of Immediate-Type Allergic Reactions

ELMER L. BECKER

I. Introduction . . . . .	267
II. Sensitization . . . . .	270
III. Components of the Allergic Reaction . . . . .	271
IV. Sites of the Antigen—Antibody Reaction . . . . .	285
V. Time Course of Allergic Reactions . . . . .	286
VI. The Terrain . . . . .	289
VII. Basis and General Description of the Classification . . . . .	291
VIII. Direct Responses (Non-mediator Determined) . . . . .	298
IX. Indirect Responses (Mediator Determined) . . . . .	299
X. Mixing of Categories in Natural Reactions . . . . .	305
XI. Pseudoallergic Reactions . . . . .	307
References . . . . .	308
AUTHOR INDEX . . . . .	315
SUBJECT INDEX . . . . .	333

# Structure and Function of Human Immunoglobulin E

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I. Introduction	1
II. Isolation and Physicochemical Characteristics of Immunoglobulin E	2
A. Identification of Myeloma Protein ND	2
B. Isolation	4
C. Chemical and Physical Characteristics	15
III. Properties of Antigenically and Biologically Active Structural Regions of Immunoglobulin E	19
A. Properties of $\epsilon$ Chains	19
B. Properties of Enzymatic Fragments	20
IV. Methods for Determination	28
V. Levels of Immunoglobulin E in Healthy Individuals	29
A. Levels in Serum	29
B. Levels in Secretions	32
VI. Levels of Immunoglobulin E in Disease	35
A. Levels in Serum	35
B. Levels in Secretions	43
C. Factors Influencing Immunoglobulin E Level	44
VII. Detection of Antibody Activity in the Immunoglobulin E Class	45
A. Red Cell-Linked Antigen-Antiglobulin Reaction	45
B. The Radioallergosorbent Test	46
C. Allergen Antibodies in Serum	48
VIII. Metabolism	49
IX. Concluding Remarks	51
References	51

## I. Introduction

Immunoglobulin E (IgE) represents a minor but distinct class of proteins present in serum of man and higher primates and possibly also in the serum of other species. In healthy individuals, the upper range of concentration is usually below 1  $\mu\text{g./ml.}$  The detection and quantitation of IgE require very sensitive methods. Immunoglobulin E is elevated 4-30 times normal in various diseases, among which atopic disorders and parasitic infestations appear to be the most prominent. Pathological amounts of IgE have also been found in the serum of patients with  $\gamma\text{E}$  myeloma.

The association of certain reaginic antibodies to a new class of im-

munoglobulin was postulated by K. Ishizaka and co-workers (1966a,b). The discovery that the myeloma protein ND and its normal counterpart share antigenic characteristics with reaginic antibodies and, in addition, carry skin-fixing structures, which appear similar to those of reagins, opened new possibilities to study the immunological and structural features of immediate hypersensitive reaction. The aim of this paper is to summarize our present knowledge of the biological and structural properties of IgE and its occurrence in various body fluids in health and disease. To this end, particular emphasis has been given to the methodology of identification and quantitation and also to the problem of isolation and characterization of IgE. References to the massive literature on reagins will be made only when found to be relevant for the understanding of a particular problem, and no attempts have been made to portray the long history of reagins, since this has been so masterly done in previous reviews by several authors (see K. Ishizaka and Ishizaka, 1968a; Sehon and Gyenes, 1965; Stanworth, 1963).

## II. Isolation and Physicochemical Characteristics of Immunoglobulin E

As a result of the obvious difficulties encountered in the isolation of reasonably homogeneous samples of a protein, which, like IgE, represents only a minor serum constituent, the physicochemical characteristics given in this paper will refer mainly to the first described E myeloma protein, ND. However, there is sufficient experimental evidence now available to support the belief that the ND protein has its major biological, immunological, and physicochemical characteristics in common with the IgE present in normal serum (Bennich *et al.*, 1968).

### A. IDENTIFICATION OF MYELOMA PROTEIN ND

Our first attempt in 1965 to isolate the atypical myeloma protein ND was done by zone electrophoresis (Johansson and Bennich, 1967a). The M component migrated in the fast  $\gamma$  region. The isolated fraction, containing 93% of the M component contaminated mainly with IgG, was used for the first immunization experiments in rabbits and for carbohydrate analysis. The latter indicated that the M-component contained about 10% of total carbohydrate—a result suggesting a possible relationship of ND to IgA or IgM. Gel filtration experiments on serum ND on calibrated columns of Sephadex G-150 gave results in the same direction; indications that the M component distributed within the same elution volume as monomeric or 7S IgA initiated a direct comparison of a monomeric A myeloma protein and protein ND. Both proteins were isolated from serum by precipitation with sodium sulfate and subsequently purified by recycling chromatography on Sephadex G-150 to



eliminate contaminating IgG. The purified A and ND proteins were added to a solution of monomeric normal IgG and the mixture was analyzed on a calibrated column of Sephadex G-200. The distribution of IgA and protein ND coincided completely as determined by quantitative immunological analysis, and the elution volume was significantly smaller than that of IgG. In contrast to these results, ultracentrifugal analyses indicated a significant difference for IgA and protein ND, the sedimentation constant values ( $s_{20,w}^0$ ) were 6.5 and 7.9, respectively.

Molecular weight determinations gave a value of about 139,000 for IgA and 196,000 for protein ND using a partial specific volume of 0.713 for both proteins. By reduction of protein ND with mercaptan followed by dissociation in acid, about 20% of the protein moiety could be recovered as  $\lambda$  chains. The remaining 80% constituted a single carbohydrate-containing component with a characteristic electrophoretic mobility in starch gel electrophoresis in acid urea. This major constituent was regarded as representing the heavy chain of an atypical immunoglobulin.

The problem of preparing class-specific antisera to IgE(ND) was not solved until fragments of ND protein were isolated (see Section III,B). Hereby it also became possible to develop the radioimmunosorbent test (RIST) described in Section IV and the radioallergosorbent test (RAST) described in Section VII. By using the RIST, a counterpart to ND was found in normal serum. The concentration in healthy individuals are usually found to be extremely low as will be further discussed in Section V. However, by chance the serum from one of the apparently healthy blood donors included in the first series of experiments was found to contain a significantly higher level of IgE(ND) than the main level of the test group. The donor was subsequently found to have a previously clinically undiagnosed hypersensitivity to dog dander, a finding which initiated a study of the level of IgE(ND) in patients suffering from asthma and hay fever, as will be further discussed in Section VI. The significantly higher level of IgE(ND) found in cases of extrinsic asthma strongly suggested a relation to reagenic antibodies as did the presence of allergen antibodies of IgE class in these patients.

In 1966, K. Ishizaka *et al.* (1966a), from their studies on antiragweed antibodies in reagin-containing fractions of atopic sera, suggested the presence of a unique immunoglobulin as a carrier of reagenic activity. The specific activity was found in the  $\gamma_1$  region by radioimmuno-electrophoresis and the protein was tentatively designated  $\gamma$ E-globulin. An exchange of antisera between Denver and Uppsala was made in March 1967 and, by comparatively antigenic analyses of myeloma protein ND and  $\gamma$ E-globulin, direct immunological evidence was obtained that