Laboratory techniques in rabies

Fourth edition

Edited by

F.-X. Meslin
M. M. Kaplan
H. Koprowski

World Health Organization
Geneva

Laboratory techniques in rabies

FOURTH EDITION

Edited by

F.-X. Meslin
Chief
Veterinary Public Health
Division of Communicable Diseases
World Health Organization
Geneva, Switzerland

M. M. Kaplan
Former Director
Research Promotion and Development
World Health Organization
Geneva, Switzerland

H. Koprowski
Director
Center for Neurovirology
Jefferson Cancer Institute
Thomas Jefferson University
Philadelphia
PA, USA

World Health Organization
Geneva
1996

WHO Library Cataloguing in Publication Data

Laboratory techniques in rabies / edited by F.-X. Meslin, M. M. Kaplan, H. Koprowski. — 4th ed.

1.Rabies *diagnosis* *laboratory manuals*
2.Rabies vaccine I.Meslin, F.-X. II.Kaplan, M.M. III.Koprowski, H.

ISBN 92 4 154479 1 *(NLM Classification: WC 550)*

Typeset in India
Printed in Finland
93/9814–Macmillans/Vammala–8500

Preface

During the 20 years that have elapsed since the publication of the third edition of *Laboratory techniques in rabies*, enormous progress has been made in improving methods of rabies vaccine and antisera production, and in developing new diagnostic and assay procedures. Major advances in molecular biology techniques have been extensively applied to the study of the rabies virus during recent years, and a fourth edition of the monograph has therefore become necessary. This edition includes some 30 new chapters, which describe new diagnostic, research and vaccine production techniques. Although some of these methods are currently restricted to relatively advanced laboratories (e.g. monoclonal antibody techniques, the polymerase chain reaction and virus expression systems), they are expected to become routine procedures in the future. Nevertheless, many laboratories will not have the facilities or equipment to use these methods, therefore the basic classical techniques described in the previous edition have been retained and, where necessary, brought up to date.

The production of rabies vaccines for animal and human use is extensively reviewed. The production of modified live-virus vaccines and recombinant vaccines is also briefly covered. It should be noted that there has been a dramatic increase in the number of cell-culture vaccines available for human use and that production is no longer restricted to developed countries. Many of these vaccines have now replaced those derived from nerve tissue. Accordingly, only two chapters deal with the production of the latter, which are still used in some developing countries.

It should be stressed that claims for the efficacy of particular vaccines are entirely the responsibility of the authors, and that their inclusion in this book does not imply official recognition by WHO. Vaccine manufacturers intending to use the production techniques described here should refer to the requirements for rabies vaccines for human and veterinary use, as defined by the WHO Expert Committee on Biological Standardization.[1-3]

An early draft manuscript of this fourth edition of *Laboratory techniques in rabies* was examined by the WHO Expert Committee on Rabies in September 1991,[4] and a number of suggestions were made for changes to the text and for the

[1] *WHO Expert Committee on Biological Standardization. Thirty-first report.* Geneva, World Health Organization, 1981 (WHO Technical Report Series. No. 658), Annex 2; Annex 3.

[2] *WHO Expert Committee on Biological Standardization. Thirty-seventh report.* Geneva, World Health Organization, 1987 (WHO Technical Report Series, No. 760), Annex 9.

[3] *WHO Expert Committee on Biological Standardization. Forty-third report.* Geneva, World Health Organization, 1994 (WHO Technical Report Series, No. 840), Annex 4; Annex 5; Annex 6.

[4] *WHO Expert Committee on Rabies. Eighth report.* Geneva, World Health Organization, 1992 (WHO Technical Report Series, No. 824).

inclusion of some additional material. In preparing the final manuscript, the editors have ensured that the information is as up to date as possible. Where new material could not be incorporated in the existing text, it has been added in the form of appendices at the end of the book.

The World Health Organization gratefully acknowledges the collaboration of the many eminent specialists who have contributed to this volume. The editors thank Miss C. Allsopp, Office of Publications, WHO, for her assistance in the preparation of this book.

List of acronyms and abbreviations used in this book

ABT	antibody-binding test
ATCC	American Type Culture Collection
BHK	baby hamster kidney cells
BPL	beta-propiolactone
BSA	bovine serum albumin
CDC	Centers for Disease Control and Prevention (USA)
cDNA	complementary deoxyribonucleic acid
C-ELISA	competitive enzyme-linked immunosorbent assay
CER	chick embryo-related cells
CI_{95}	95% confidence interval
CVS	Challenge Virus Standard
DI	defective interfering (particles)
DIA	dot-immunobinding assay
DMEM	Dulbecco's modified Eagle's medium
DNA	deoxyribonucleic acid
EAE	experimental allergic encephalomyelitis
EBL	European bat lyssavirus
EBM	Eagle's basal medium
ED_{50}	median effective dose, 50% end-point dilution
EIA	enzyme immunoassay
ELISA	enzyme-linked immunosorbent assay
EMEM	Eagle's minimum essential medium
ERA	Evelyn Rokitniki Abelseth strain of rabies virus
ERIG	equine rabies immunoglobulin
FA	fluorescent antibody
FCS	fetal calf serum
FDA	Food and Drug Administration (USA)
FFD_{50}	dilution at which 50% of the observed microscopic fields contain one or more foci of infected cells
FFU	focus-forming units
FITC	fluorescein isothiocyanate
FRhMDC	fetal rhesus monkey diploid cell
FWR	French wild rabies isolates
G protein	rabies glycoprotein
HDC	human diploid cell
HEP	Flury high egg passage strain of rabies virus
HN	haemagglutinin–neuraminidase protein
HRIG	human rabies immunoglobulin

Ig	immunoglobulin
INHV	infectious haematopoietic necrosis virus
INPPAZ	PAHO/WHO Pan American Institute for Food Protection and Zoonoses (Argentina)
IU	International Unit
LD_{50}	median lethal dose
LEP	Flury low egg passage strain of rabies virus
L protein	rabies RNA-dependent RNA polymerase
M1 protein	rabies phosphoprotein
M2 protein	rabies matrix or membrane protein
MAb	monoclonal antibody
MAb-G	anti-glycoprotein monoclonal antibody
MAb-N	anti-nucleoprotein monoclonal antibody
MAb-RNP	anti-ribonucleoprotein monoclonal antibody
MEM	minimum essential medium
$MICLD_{50}$	median lethal dose for mice inoculated by the intracerebral route
MIT	mouse inoculation test
MLV	modified live-virus
MNA	mouse neuroblastoma cells
MNT	mouse neutralization test
MOI	multiplicity of infection
Mok	Mokola virus
mRNA	messenger ribonucleic acid
NA	neuroblastoma cells
NIH	National Institutes of Health (USA)
N protein	rabies nucleoprotein
OD	optical density
PAb	polyclonal antibody
PAb-G	anti-glycoprotein polyclonal antibody
PAHO	Pan American Health Organization
PAS	Louis Pasteur strain of rabies virus
PBS	phosphate-buffered saline
PCEC	purified chick embryo cell
PCR	polymerase chain reaction
PDE	purified duck embryo
PDL	population doubling level
PFU	plaque-forming units
PHKC	primary Syrian hamster kidney cell
PM	Pitman-Moore strain of rabies virus
PSRV	product-specific reference vaccine
PV	Pasteur strain of rabies virus
PVRV	purified Vero cell rabies vaccine
RIG	rabies immunoglobulin
RNA	ribonucleic acid
RNP protein	rabies ribonucleoprotein
RTCIT	rabies tissue-culture infection test
RFFIT	rapid fluorescent focus inhibition test
RREID	rapid rabies enzyme immunodiagnosis
SAD	Street-Alabama–Dufferin strain of rabies virus

SCID	severe combined immunodeficient
SDS–PAGE	sodium dodecyl sulfate–polyacrylamide gel electrophoresis
SMB	suckling mouse brain
$TCID_{50}$	median tissue-culture infective dose
VRG	recombinant vaccinia virus expressing the G protein gene of rabies virus
VSV	vesicular stomatitis virus

Contents