

TABLE OF CONTENTS

Chapter 1

WATERBORNE VIRUSES	1
1.1. THE CLASSIFICATION OF VIRUSES IN ENVIRONMENTAL WATERS	2
1.1.1. Picornaviridae	2
1.1.2. Reoviridae	5
1.1.3. Coronaviridae	6
1.1.4. Caliciviridae	7
1.1.5. Astroviridae	7
1.1.6. Adenoviridae	7
1.1.7. Norwalk and Similar Viruses	8
1.1.8. Non-A and Non-B Hepatitis Viruses	9
1.2. VIRAL CONTAMINATION OF WATER	9
1.2.1. Viral Contamination of Wastewater	9
1.2.2. Viral Contamination of Recreational Waters	12
1.2.3. Viral Contamination of Water Distribution Systems	13
1.3. EPIDEMIOLOGY	13
1.3.1. The Accepted Epidemiological Facts	14
1.3.2. The Minimal Infecting Dose	17
REFERENCES	18

Chapter 2

GATHERING, MONITORING, AND STORAGE OF SAMPLES	23
2.1. SAMPLING MATERIAL	23
2.1.1. Preparation of the Material	23
2.1.2. Sterilization of the Material	24
2.1.3. Verification of the Sterilization	24
2.2. CONTROL OF THE SAMPLING PERSONNEL AND TECHNICIANS	26
2.3. SAMPLING DESIGN FOR MONITORING WATERS	27
2.3.1. Sampling of Wastewater	28
2.3.2. Sampling of River Water	28
2.3.3. Sampling of Potable Water	30
2.4. STORAGE OF SAMPLES	31
REFERENCES	32

Chapter 3	
CONCENTRATION METHODS	33
3.1. THE GAUZE PAD METHOD	36
3.1.1. Preparation of the Gauze Pads	36
3.1.2. Positioning of the Gauze Pads	37
3.1.3. Recovery of the Pads and Water	37
3.1.4. Limitations of the Method	37
3.2. ADSORPTION-ELUTION METHODS	37
3.2.1. Cellulose Ester Filters	37
3.2.2. Glass Microfiber Filters	38
3.2.3. Filtration on Powdered Glass	40
3.2.4. Positively Charged Filters	43
3.3. SECONDARY CONCENTRATION METHODS	44
3.3.1. Secondary Concentration by Organic Flocculation	44
3.3.2. Secondary Concentration with Aluminum Hydroxide	45
3.3.3. Secondary Concentration by Adsorption-Elution on Iron Oxide	46
3.4. DECONTAMINATION OF THE SAMPLES	47
3.4.1. Treatment with Chloroform	47
3.4.2. Treatment with Antibiotics	47
3.5. DETOXIFICATION OF THE SAMPLES	47
REFERENCES	48

Chapter 4	
SYSTEMS FOR THE DETECTION OF VIRUSES	51
4.1. CELL CULTURES	51
4.1.1. The Vessels for Culture	52
4.1.2. Culture Media	58
4.1.3. The Cells	61
4.2. ANIMALS	66
REFERENCES	66

Chapter 5	
ISOLATION TECHNIQUES	69
5.1. ISOLATION ON CELL CULTURES	69
5.1.1. Choice of Cell Systems	69
5.1.2. Isolation on Integral Cellular Layers	69
5.1.3. Isolation Using a Cell Suspension	72
5.1.4. Limitations of the Isolation Techniques on Cell Cultures	75
5.2. ISOLATION BY INOCULATION OF MICE	75
5.2.1. Inoculation	75
5.2.2. Observation	76
REFERENCES	76

Chapter 6

IDENTIFICATION TECHNIQUES	77
6.1. IDENTIFICATION OF VIRUSES PROVOKING A CYTOPATHIC EFFECT ON CELL CULTURES	77
6.1.1. Preliminary Diagnosis	77
6.1.2. Immunological Identification by Neutralization	80
6.1.3. Intratypic Differentiation of Poliomyelitic Viruses	83
6.2. IDENTIFICATION OF THE VIRUSES MULTIPLYING ON CELL CULTURES WITHOUT PROVOKING A CYTOPATHIC EFFECT	87
6.2.1. Detection of Rotaviruses	87
6.3. IDENTIFICATION OF VIRUSES ISOLATED FROM NEWBORN MICE	88
6.3.1. Histological Examination	88
6.4. IDENTIFICATION WITHOUT INOCULATION INTO SUSCEPTIBLE SYSTEMS	89
6.4.1. Electron Microscopy and Immunomicroscopic Methods	89
6.4.2. The SPACE Test	89
6.4.3. Immunoenzymological Methods	90
REFERENCES	92

Chapter 7

METHODS FOR QUANTIFYING RESULTS	95
7.1. DILUTION OF THE SAMPLES	96
7.2. ENUMERATIVE METHODS	96
7.2.1. Fewer Than 15 Plaques at One Level of Dilution	98
7.2.2. Number of Plaques Counted Between 15 and 100 at One Level of Dilution	99
7.2.3. More Than 100 Plaques at One Level of Dilution	99
7.2.4. Estimation of the Viral Density from Results Taken at Several Levels of Dilution	99
7.2.5. Comparison of the Results	102
7.3. QUANTAL NUMERATIVE METHODS	102
7.3.1. The 50% Cytopathic dose (CPD ₅₀)	102
7.3.2. The Most Probable Number of Cytopathic Units (MPNCU)	105
7.4. CHOICE OF A METHOD OF QUANTIFICATION	115
REFERENCES	117

Chapter 8

ORGANIZATION OF A VIROLOGY LABORATORY FOR WATER SYSTEMS	119
8.1. DEFINITIONS OF WORK UNITS	119
8.1.1. Unit for Waste, Surface, and Recreational Waters	119
8.1.2. Work Unit for Potable Water	119

8.1.3. Unit for Cell Cultures	119
8.1.4. Isolation and Identification Unit	120
8.1.5. Cleaning and Sterilization Unit	120
8.2. EQUIPMENT FOR THE WORK UNITS	120
8.3. MAINTENANCE OF THE WORK UNITS	121
8.4. ORGANIZATION OF THE WORK UNITS	122
8.4.1. Asepsis of the Manipulations	122
8.4.2. Protection of the Handler	123
REFERENCES	124

Chapter 9

CONDUCTING A VIROLOGICAL ANALYSIS	125
9.1. SAMPLING OF WASTE WATERS	125
9.1.1. Method of Sampling	125
9.1.2. Volume of the Sample	126
9.1.3. Transport of the Sample	127
9.2. SAMPLING OF SURFACE WATERS	127
9.2.1. Method of Sampling	127
9.2.2. Volume of the Sample	127
9.2.3. Transport of the Sample	127
9.3. SAMPLING POTABLE WATER	128
9.3.1. Method of Sampling	128
9.3.2. Transport of the Sample	128
9.4. CONCENTRATION OF THE SAMPLES	128
9.4.1. Very Turbid Water	129
9.4.2. Slightly Turbid or Nonturbid Water	129
9.5. DETECTION OF VIRUSES	129
9.5.1. Detection by Inoculation on Cell Cultures	129
9.5.2. Detection by Immunological Techniques	129
INDEX	131