



ECOLOGY OF **INVERTEBRATE DISEASES**

Edited by Ann E. Hajek and David I. Shapiro-Ilan



WILEY

Ecology of Invertebrate Diseases

Ecology of Invertebrate Diseases

Edited by

Ann E. Hajek

Cornell University, Ithaca, New York, US

David I. Shapiro-Ilan

USDA-ARS, Byron, Georgia, US

WILEY

This edition first published 2018
© 2018 John Wiley & Sons Ltd.

All rights reserved. No part of this publication may be reproduced, stored in a retrieval system, or transmitted, in any form or by any means, electronic, mechanical, photocopying, recording or otherwise, except as permitted by law. Advice on how to obtain permission to reuse material from this title is available at <http://www.wiley.com/go/permissions>.

The right of Ann E. Hajek and David I. Shapiro-Ilan to be identified as the authors of the editorial material in this work has been asserted in accordance with law.

Registered Office(s)

John Wiley & Sons, Inc., 111 River Street, Hoboken, NJ 07030, USA
John Wiley & Sons Ltd, The Atrium, Southern Gate, Chichester, West Sussex, PO19 8SQ, UK

Editorial Office

9600 Garsington Road, Oxford, OX4 2DQ, UK

For details of our global editorial offices, customer services, and more information about Wiley products visit us at www.wiley.com.

Wiley also publishes its books in a variety of electronic formats and by print-on-demand. Some content that appears in standard print versions of this book may not be available in other formats.

Limit of Liability/Disclaimer of Warranty

While the publisher and authors have used their best efforts in preparing this work, they make no representations or warranties with respect to the accuracy or completeness of the contents of this work and specifically disclaim all warranties, including without limitation any implied warranties of merchantability or fitness for a particular purpose. No warranty may be created or extended by sales representatives, written sales materials or promotional statements for this work. The fact that an organization, website, or product is referred to in this work as a citation and/or potential source of further information does not mean that the publisher and authors endorse the information or services the organization, website, or product may provide or recommendations it may make. This work is sold with the understanding that the publisher is not engaged in rendering professional services. The advice and strategies contained herein may not be suitable for your situation. You should consult with a specialist where appropriate. Further, readers should be aware that websites listed in this work may have changed or disappeared between when this work was written and when it is read. Neither the publisher nor authors shall be liable for any loss of profit or any other commercial damages, including but not limited to special, incidental, consequential, or other damages.

Library of Congress Cataloging-in-Publication Data

Names: Hajek, Ann E., editor. | Shapiro-Ilan, David I., editor.

Title: Ecology of invertebrate diseases / Edited by Ann E. Hajek, David I. Shapiro-Ilan.

Description: Hoboken, NJ : John Wiley & Sons, 2017. | Includes bibliographical references and index. |

Identifiers: LCCN 2017023661 (print) | LCCN 2017035570 (ebook) | ISBN 9781119256014 (pdf) | ISBN 9781119256069 (epub) | ISBN 9781119256076 (cloth)

Subjects: LCSH: Invertebrates--Ecology. | Invertebrates--Diseases.

Classification: LCC QL364.4 (ebook) | LCC QL364.4 .E26 2017 (print) | DDC 592.17/82--dc23

LC record available at <https://lcn.loc.gov/2017023661>

Cover Design: Wiley

Cover Image: Top photo by Ann E. Hajek; bottom photo by Ivan Hiltbold

Set in 10/12pt WarnockPro by SPi Global, Chennai, India

Contents

List of Contributors *xvii*

Preface *xxi*

Section I Introduction *1*

1 General Concepts in the Ecology of Invertebrate Diseases *3*

Ann E. Hajek and David I. Shapiro-Ilan

1.1 Introduction *3*

1.1.1 What Is Disease? *4*

1.1.2 Terminology and Measurements *5*

1.1.2.1 Prevalence/Incidence *5*

1.1.2.2 Pathogenicity/Virulence *5*

1.1.2.3 Infection/Infectivity *6*

1.1.2.4 Immunity *6*

1.1.2.5 Transmission *7*

1.1.2.6 Epizootic and Enzootic Diseases *7*

1.1.2.7 Cycles of Infection *8*

1.1.2.8 R_0 and the Host Density Threshold *10*

1.1.3 Factors Influencing the Ecology of Invertebrate Diseases *11*

1.1.3.1 Host Range *12*

1.2 Types of Studies *13*

1.3 Why Study the Ecology of Invertebrate Diseases? *13*

1.4 What this Book Covers *14*

Acknowledgments *16*

References *16*

2 Methods for Studying the Ecology of Invertebrate Diseases and Pathogens *19*

Raquel Campos-Herrera and Lawrence A. Lacey

2.1 Introduction *19*

2.2 Traditional Methods for Studying Diseases *19*

2.2.1 Sampling Goals *19*

2.2.2 Sampling Regimes *20*

2.2.3 Methodologies *20*

2.2.3.1 Searching for Infected Insects Using General Entomological Sampling Methods *23*

2.2.3.2	Selective Media	23
2.2.3.3	Extraction Methods	24
2.2.3.4	Airborne Spore Sampling	24
2.2.3.5	Insect Baiting	24
2.2.3.6	Dispersal of Entomopathogens: Mark–Release–Recapture Method	25
2.3	Molecular Tools to Assist in the Detection and Quantification of Pathogens and their Impact on the Host	25
2.3.1	Employment of Proteins: The Beginning of the Molecular Era in Invertebrate Pathology	26
2.3.2	Techniques Based on the Nucleic Acids: the “Pre-Omics” Era	27
2.3.3	Advanced Techniques: qPCR, NGS, and the Arrival of the -Omics Era	31
2.4	Traditional Versus Molecular Methods: Advantages and Limitations	33
2.5	Advancing the Frontiers of Ecology using Pathogens and Diseases	36
2.6	Conclusion	38
	Acknowledgments	38
	References	38

Section II The Basics of Invertebrate Pathogen Ecology 49

3 The Pathogen Population 51

Leellen F. Solter and James J. Becnel

3.1	Introduction	51
3.2	Characteristics of Pathogens	51
3.2.1	Invasiveness and Infectivity	52
3.2.1.1	Routes of Entry	53
3.2.1.2	Establishment of Infection and Tissue Tropism	54
3.2.2	Pathogenicity, Virulence, and Pathogen Replication	55
3.2.2.1	Virulence Factors	57
3.2.2.2	Attenuation or Enhancement of Virulence	58
3.2.3	Latency	59
3.2.4	Obligate, Opportunistic, and Facultative Pathogens	60
3.2.5	Transmission	61
3.2.5.1	Horizontal Transmission	61
3.2.5.2	Vertical Transmission	62
3.2.5.3	Indirect Transmission	63
3.2.6	Genetic Variability and Potential for Coevolution with Hosts	64
3.2.6.1	Species and Strains	64
3.2.6.2	Host Specificity	65
3.3	Pathogen Effects on Host Development and Behavior	66
3.4	Pathogen Populations	67
3.4.1	Density-Dependent Pathogens	67
3.4.2	Density-Independent Pathogens	68
3.4.3	Pathogen Persistence in the Host Population	68
3.4.3.1	Chronic Infections and Vertical Transmission	69
3.4.3.2	Alternative and Alternate/Intermediate Hosts	69

3.4.3.3	Pathogen Survival in Cadavers and in Plant Tissues	70
3.4.3.4	Latency in Host Populations	70
3.4.4	Persistence of Pathogen Stages in the Environment	71
3.5	Dispersal and Spatial Distribution of Pathogens	72
3.5.1	Physical Factors: Wind and Water Dispersal	73
3.5.2	Biological Factors	73
3.5.3	Spatial Distribution	74
3.6	Pathogen Interactions	75
3.6.1	Interactions with other Biological Agents	75
3.6.2	Interactions with Pesticides and Other Chemicals	77
3.6.3	Enhancing Factors	77
3.7	Conclusion	78
	References	79
4	The Host Population	101
	<i>Louela A. Castrillo</i>	
4.1	Introduction	101
4.2	General Host Factors	103
4.2.1	Routes of Pathogen Acquisition	103
4.2.2	Insect Species, Life Stage, Age, and Gender	103
4.2.3	Population Density	104
4.3	Barriers to Microbial Infection	105
4.3.1	Insect Integument	105
4.3.2	Tracheae	107
4.3.3	Insect Gut	108
4.3.3.1	Peritrophic Membrane and Basal Lamina	108
4.3.3.2	Conditions in the Gut Lumen	108
4.3.3.3	Sloughing of Infected Epidermal Cells	109
4.4	Defenses against Microbial Infection	110
4.4.1	Innate Immune System	110
4.4.1.1	Constitutive Innate Immunity: Cellular Immunity	110
4.4.1.2	Constitutive Innate Immunity: Phenoloxidase	113
4.4.1.3	Induced Innate Immunity: Reactive Oxygen Species	113
4.4.1.4	Induced Innate Immunity: Antimicrobial Peptides	114
4.4.2	Microbiome-Based Defenses	117
4.4.2.1	Gut Microbiota	117
4.4.2.2	Intracellular Symbionts	118
4.4.3	Behavioral Defenses	119
4.4.3.1	Avoidance/Evasion	120
4.4.3.2	Grooming and Hygienic Behaviors	121
4.4.3.3	Diet-Based Prophylactic and Therapeutic Defenses	122
4.4.3.4	Thermoregulation	123
4.4.3.5	Deposition of Antimicrobial Compounds	123
4.5	Resistance via Priming	124
4.6	Conclusion	125
	Acknowledgments	126
	References	126

5 Abiotic Factors 143

Dana Ment, Ikkei Shikano and Itamar Glazer

5.1 Introduction 143

5.2 The Surviving Unit 143

5.2.1 Nematodes 143

5.2.2 Fungi 144

5.2.3 Viruses 145

5.2.4 Bacteria 146

5.3 Abiotic Factors Affecting Invertebrate Pathogens 146

5.3.1 Temperature 146

5.3.1.1 Nematodes 147

5.3.1.2 Fungi 148

5.3.1.3 Viruses 150

5.3.1.4 Bacteria 152

5.3.2 Moisture and Humidity 152

5.3.2.1 Nematodes 153

5.3.2.2 Fungi 154

5.3.2.3 Viruses 155

5.3.2.4 Bacteria 156

5.3.3 Ultraviolet Radiation 156

5.3.3.1 Nematodes 157

5.3.3.2 Fungi 157

5.3.3.3 Viruses 158

5.3.3.4 Bacteria 158

5.3.4 Chemical Inputs 159

5.3.4.1 Nematodes 159

5.3.4.2 Fungi 160

5.3.4.3 Viruses 160

5.3.4.4 Bacteria 161

5.3.5 Other Habitat Characteristics 162

5.3.5.1 Nematodes 162

5.3.5.2 Fungi 163

5.3.5.3 Viruses 164

5.3.5.4 Bacteria 165

5.4 Mechanisms of Survival 165

5.4.1 Nematodes 165

5.4.2 Fungi 166

5.4.3 Viruses 166

5.4.4 Bacteria 167

5.5 Conclusion 167

References 169

6 The Biotic Environment 187

Jenny S. Cory and Pauline S. Deschodt

6.1 Introduction 187

6.2 Tritrophic Interactions 188

6.2.1 Further Complexity 190

6.3	Pathogen–Natural Enemy Interactions	191
6.3.1	Entomopathogen–Entomopathogen Interactions	191
6.3.2	Entomopathogen–Parasitoid Interactions	195
6.3.2.1	Effects of Pathogens on Parasitoids	195
6.3.2.2	Effects of Parasitoids on Pathogens	197
6.3.2.3	Population Level Effects	198
6.3.3	Pathogen–Predator Interactions	199
6.3.4	Conclusion	200
6.4	Microbe-Mediated Defense	200
6.4.1	Heritable Symbionts	201
6.4.2	Do Gut Microflora Influence Pathogen Susceptibility?	202
6.4.3	Future Directions	204
6.5	Conclusion	204
	Acknowledgments	204
	References	205

Section III Ecology of Pathogen Groups 213

7	Viruses	215
	<i>Trevor Williams</i>	
7.1	Introduction	215
7.2	Diversity of Invertebrate Pathogenic Viruses	216
7.3	Distribution of Invertebrate Pathogenic Viruses	219
7.4	Key Aspects of Pathogen Ecology	220
7.5	Transmission	221
7.5.1	Horizontal Transmission	221
7.5.1.1	Estimating Horizontal Transmission	223
7.5.2	Vertical Transmission	223
7.6	Persistence	225
7.6.1	Persistence within the Host	225
7.6.2	Persistence Outside of the Host	226
7.6.2.1	Persistence on Plants	227
7.6.2.2	Persistence in Soil	229
7.6.2.3	Persistence in Water	230
7.7	Dispersal	231
7.7.1	Host-Mediated Dispersal	231
7.7.2	Environmental Factors Involved in Dispersal	232
7.7.3	Biotic Factors that Assist the Dispersal of Viruses	233
7.7.3.1	Predators	233
7.7.3.2	Parasitoids	234
7.7.3.3	Other Organisms	234
7.7.4	Agricultural Practices that Affect Dispersal	234
7.7.5	Spatial Patterns of Dispersal	235
7.8	Genetic Diversity in Viruses	235
7.8.1	Genetic Diversity is Pervasive in Virus Populations	235
7.8.2	Genetic Diversity Favors Virus Survival	237

Contents

7.8.3	What Generates So Much Genetic Diversity?	238
7.8.4	How Is Genetic Diversity Transmitted?	239
7.9	Role of Host Behavior in Virus Ecology	240
7.9.1	Foraging Decisions: What and Where to Eat	240
7.9.2	The Risks of Cannibalism	241
7.9.3	Sexually Transmitted Viral Diseases	241
7.9.4	Ecological Consequences of Host Manipulation by Viruses	242
7.9.4.1	Molecular Basis for Host Manipulation	243
7.10	Dynamics of Viruses in Host Populations	244
7.10.1	Pathogenic Viruses Can Regulate Populations	244
7.10.2	Ecosystem Characteristics that Favor Virus Transmission	246
7.10.3	Climate Change and Insect–Virus Population Dynamics	247
7.11	Influence of Abiotic Factors on Viruses	248
7.11.1	Effect of Ultraviolet Light on Viruses	248
7.11.2	Seasonal Effects on Viruses	250
7.11.3	Effect of Temperature on Viruses	250
7.11.4	Humidity, Moisture and Precipitation	251
7.11.5	Effect of pH on Viruses	251
7.12	Biotic Factors that Interact with Virus Populations	253
7.12.1	Plant Phenology, Structure, and Nutritional Value	253
7.12.2	Phytochemical–Virus Interactions	253
7.12.3	Virus Interactions with Alternative Hosts	254
7.12.4	Competition and Facilitation in Virus Interactions with Other Organisms	255
7.12.4.1	Virus Interactions with Parasitoids	255
7.12.4.2	Virus Interactions with Other Pathogens	257
7.12.4.3	Virus Interactions with Microbiota	258
7.13	Conclusion	258
	Acknowledgments	259
	References	259
8	Bacteria	287
	<i>Trevor A. Jackson, Colin Berry and Maureen O'Callaghan</i>	
8.1	Introduction	287
8.2	Bacterial Pathogens and Associations with Insects	288
8.3	Pathogenicity and Virulence	294
8.3.1	Pathogenicity	295
8.3.2	Virulence	299
8.4	Disease Transmission	300
8.5	Survival in the Environment	301
8.5.1	Soil	302
8.5.2	Aqueous Environments	304
8.5.3	On the Phylloplane and <i>In Planta</i>	304
8.6	Population Dynamics: Epizootics and Enzootics	305
8.7	Evolution	308
8.8	Ecology Guiding Use of Bacterial Entomopathogens in Microbial Control	309

8.9	Conclusion	311
	References	312
9	Fungi	327
	<i>Ann E. Hajek and Nicolai V. Meyling</i>	
9.1	Introduction	327
9.1.1	Fungal Systematics and Taxonomy	328
9.1.2	Relevance of Fungal Systematics and Taxonomy in Ecology	330
9.2	Fungal Biology and Pathology	331
9.2.1	Biology and Pathology of Major Groups of Fungal Pathogens	331
9.2.1.1	Entomophthoromycotina, Entomophthorales	331
9.2.1.2	Ascomycota, Hypocreales	333
9.2.2	Distribution Patterns and Habitat Associations of Invertebrate Pathogenic Fungi	334
9.2.2.1	Patterns of Fungal Abundance and Distribution: Insights from the Use of Molecular Markers	335
9.2.3	Factors Governing Diversity Patterns of Fungal Pathogens	337
9.3	Dynamics of Fungal Pathogens	338
9.3.1	Disease Transmission	338
9.3.2	Fungal Dispersal	341
9.3.3	Fungal Environmental Survival and Persistence	341
9.3.4	Impacts on Host Population Densities over Space and Time	342
9.4	Interactions between Fungal Pathogens and Host Individuals	344
9.4.1	Host Responses to Fungal Pathogens to Prevent or Cure Infections	344
9.5	Impact of Abiotic Factors on Infected Hosts and Pathogen Inocula	347
9.6	Impact of Biotic Factors on Pathogenic Fungi	349
9.6.1	Endophytic and Rhizosphere Associations of Invertebrate Fungal Pathogens	349
9.6.1.1	Natural Occurrence and Distribution of Invertebrate Pathogenic Fungi as Plant Associates	350
9.6.1.2	Experimental Inoculations of Plants with Entomopathogenic Fungi	351
9.6.1.3	Direct and Indirect Fungal Interactions with Insects and Plants	352
9.6.2	Interactions between Host Symbionts and Fungal Pathogens	353
9.6.3	Interactions between Fungal Pathogens and Other Natural Enemies	354
9.6.3.1	Interactions among Co-infecting Pathogens	355
9.6.3.2	Interactions of Fungal Pathogens with Parasitoids and Predators	356
9.6.4	Mycoparasitism of Fungal Pathogens	357
9.7	Use of Pathogenic Fungi for Biological Control of Invertebrates	358
9.8	Conclusion	361
	Acknowledgments	361
	References	362

10 Microsporidia 379

Gernot Hoch and Leellen F. Solter

- 10.1 Introduction 379
 - 10.1.1 Mechanisms of Infection 380
 - 10.1.2 Microsporidian Life Cycles 381
 - 10.1.3 Pathology 381
- 10.2 Host Population 383
 - 10.2.1 Susceptibility to Microsporidiosis 383
 - 10.2.2 Immune Response 383
 - 10.2.3 Behavioral Response 384
- 10.3 Pathogen Population 385
 - 10.3.1 Virulence of Microsporidian Pathogens 385
 - 10.3.2 Host Specificity 386
 - 10.3.2.1 Physiological vs. Ecological Host Specificity 386
 - 10.3.2.2 Host Range 386
 - 10.3.2.3 Alternate Hosts 387
 - 10.3.2.4 Microsporidia Crossing the Invertebrate–Vertebrate Barrier 387
 - 10.3.3 Persistence in the Environment 388
- 10.4 Transmission 390
 - 10.4.1 Horizontal Transmission 390
 - 10.4.1.1 Transmission from Living Hosts 390
 - 10.4.1.2 Transmission after Host Death 392
 - 10.4.1.3 Transmission by Parasitoid Vectors 393
 - 10.4.1.4 Effects of Host Development and Host–Microsporidia Interactions on Transmission 393
 - 10.4.2 Vertical Transmission 394
- 10.5 Epizootiology 397
 - 10.5.1 Microsporidian Prevalence in Invertebrate Populations and Impact on Host Populations 397
 - 10.5.2 Microsporidia in Cultured Insects 398
 - 10.5.3 Microsporidia as Potential Biological Control Agents 399
- References 400

11 Nematodes 415

David I. Shapiro-Ilan, Ivan Hiltbold and Edwin E. Lewis

- 11.1 Introduction 415
 - 11.1.1 Diversity and Life Histories 415
 - 11.1.2 EPN Distribution 420
- 11.2 Transmission 421
- 11.3 Host Population 421
- 11.4 Pathogen Population 422
 - 11.4.1 Pathogenicity and Virulence 422
 - 11.4.2 Persistence and Recycling 423
 - 11.4.3 Dispersal and Foraging Behavior 423
- 11.5 Abiotic Environmental Factors 424
 - 11.5.1 Soil Moisture 424

11.5.2	Soil Temperature	425
11.5.3	Soil Characteristics and Chemistry	425
11.5.4	Ultraviolet Light	426
11.6	Biotic Interactions	426
11.6.1	Interactions with Predators and Pathogens, Including Intraguild Competition	426
11.6.2	Cues Used in Host-Finding and Navigation	427
11.6.3	Tri-trophic Interactions (Plant, Insect, Nematode)	427
11.7	Applied Ecology and Aspects in Microbial Control	427
11.7.1	Production, Formulation, and Application	427
11.7.2	Approaches to Microbial Control	428
11.8	Conclusion	430
	References	431
 Section IV Applied Ecology of Invertebrate Pathogens 441		
12	Modeling Insect Epizootics and their Population-Level Consequences	443
	<i>Bret D. Elder</i>	
12.1	Introduction	443
12.2	The Pathogen and its Hosts	445
12.3	Modeling Disease Transmission: A Single Epizootic	447
12.3.1	Phenomenological and Mechanistic Models	448
12.4	Fitting Models to Data	450
12.4.1	Akaike Information Criterion	451
12.4.2	An Example of the AIC in Action	452
12.5	A Bayesian Approach	453
12.5.1	Fitting a Bayesian Model	454
12.5.2	An Example of the WAIC in Action	456
12.6	Long-Term Dynamics	457
12.6.1	Long-Term Dynamics: Confronting Models with Data	458
12.6.2	Time-Series Diagnostics	459
12.7	Modifying and Applying the Model	462
12.8	Conclusion	463
	Acknowledgments	463
	References	463
 13	 Leveraging the Ecology of Invertebrate Pathogens in Microbial Control	 469
	<i>Surendra K. Dara, Tarryn A. Goble and David I. Shapiro-Ilan</i>	
13.1	Basics of Microbial Control and Approaches	469
13.1.1	Classical Microbial Control	469
13.1.2	Inoculative Release	471
13.1.3	Inundative Release	472
13.1.4	Conservation/Environmental Manipulation	472
13.2	Ecological Considerations	472
13.2.1	Host Specificity	472
13.2.2	Dispersal Ability	473
13.2.3	Virulence	473

Contents

13.2.4	Pathogen Density	473
13.2.5	Host-Related Factors	474
13.2.6	Transmission	474
13.2.7	Environmental Persistence	475
13.3	Methods to Improve Microbial Control	476
13.3.1	Improving the Organism as a Microbial Control Agent	476
13.3.2	Improving Production Methods	477
13.3.3	Improving Formulation and Application Technologies	478
13.3.4	Improving the Environment	479
13.4	Incorporating Microbial Control into Integrated Pest-Management Systems	480
13.4.1	Regulatory Issues	480
13.4.2	Standalone vs. Integrated Approaches	481
13.4.3	Case Studies	481
13.4.3.1	Orchard Crops	482
13.4.3.2	Row Crops	482
13.4.3.3	Forests	483
13.4.3.4	Greenhouses	483
13.5	Conclusion	484
	References	484
14	Prevention and Management of Diseases in Terrestrial Invertebrates	495
	<i>Jørgen Eilenberg and Annette Bruun Jensen</i>	
14.1	Introduction	495
14.1.1	Types of Production Facilities	496
14.1.2	Transmission of Insect Diseases in Production Facilities	499
14.2	Major uses of Insects and Mites in the Production and Transmission of Insect Pathogens within Production Systems	500
14.2.1	Pollination and Honey Production	500
14.2.2	Silk Production	507
14.2.3	Biological Control	509
14.2.4	Production of Insects for Food and Feed	512
14.3	Status of Diagnostic Services	516
14.4	Ensuring Production of Healthy Insects	516
14.5	Conclusion	519
	Acknowledgments	519
	References	519
15	Prevention and Management of Infectious Diseases in Aquatic Invertebrates	527
	<i>Jeffrey D. Shields</i>	
15.1	Scope	527
15.1.1	Myriad Pathogens Infect Aquatic Invertebrates	527
15.1.2	Overview of Disease Issues in Assessing Epidemics in Aquatic Invertebrates	531
15.2	Oyster Diseases	539
15.3	Crustacean Diseases	543
15.3.1	Outbreaks in Shrimp Aquaculture	544

15.3.2	Disease Management in Shrimp Aquaculture	548
15.3.2.1	Switching Species and Specific Pathogen-Free (SPF) Stocks	549
15.3.2.2	Surveillance	550
15.3.2.3	Development of “Vaccines”	551
15.3.2.4	Ecological and Biological Control	551
15.3.3	Crayfish and Krebspest	552
15.3.4	Disease Emergence in Culture of the Chinese Mitten Crab	553
15.4	Crustacean Fisheries	554
15.4.1	Snow Crabs and Bitter Crab Disease	555
15.4.2	American Lobster and Epizootic Shell Disease	556
15.4.3	Spiny Lobsters and PaV1	559
15.5	Agencies for Disease Management	560
15.6	Conclusion	563
	Acknowledgments	563
	References	563
16	Ecology of Emerging Infectious Diseases of Invertebrates	587
	<i>Colleen A. Burge, Amanda Shore-Maggio and Natalie D. Rivlin</i>	
16.1	Introduction	587
16.2	Host–Pathogen Relationships and Anthropogenic Change	593
16.2.1	Ecological Context of Invertebrate Host–Pathogen Relationships	593
16.2.2	Anthropogenic Change and Disease Emergence	594
16.2.2.1	Host Factors	595
16.2.2.2	Pathogen Factors	595
16.2.2.3	Environment Factors	595
16.3	Case Studies of Invertebrate Disease Emergence	596
16.3.1	Molluscan Herpesvirus Infections of Bivalves	597
16.3.1.1	OsHV-1 Infections of Pacific Oysters and Other Bivalves	598
16.3.2	Acute Hepatopancreatic Necrosis Disease of Shrimp	601
16.3.3	Emerging Densoviruses of Arthropods and Echinoderms	603
16.3.3.1	Acheta Domesticus Densovirus	603
16.3.3.2	Sea Star-Associated Densovirus	604
16.3.4	Emerging Pathogens of Pollinators	605
16.3.4.1	<i>Varroa destructor</i> and Deformed Wing Virus	606
16.3.4.2	Spillover and Spread of <i>Nosema ceranae</i>	607
16.3.4.3	Multi-stressors, Bee Mortalities and Control Measures	608
16.3.5	Emergent Coral Diseases	608
16.3.5.1	Black Band Disease	609
16.3.5.2	Acroporid Serratosis	610
16.3.5.3	Problems Facing Coral Disease Investigations	610
16.4	Conclusion	611
	Acknowledgments	612
	References	612

17	Conclusions and Future Directions	627
	<i>David Shapiro-Ilan and Ann E. Hajek</i>	
17.1	The Increasing Urgency of the Study of Invertebrate Pathogen Ecology	627
17.1.1	Food Security and the Role of Microbial Control	627
17.1.2	Conservation of Beneficial Organisms	628
17.2	The Future for Invasive and Native Invertebrate Pathogens	629
17.3	New Directions and Novel Tools for Studying Invertebrate Ecology	630
17.3.1	Molecular Tools	630
17.3.2	Chemical Ecology and Signaling	631
17.3.3	Exploring Other Novel Biotic Associations	632
17.3.4	Interdisciplinary Studies	633
	References	634
	Index	637

List of Contributors

James J. Becnel

USDA ARS CMAVE
Gainesville, FL, USA

Colin Berry

Cardiff School of Biosciences
Cardiff University
Cardiff, UK

Colleen A. Burge

Institute of Marine and Environmental
Technology
University of Maryland Baltimore
County
Baltimore, MD, USA

Raquel Campos-Herrera

MeditBio
University of Algarve
Faro, Portugal

Louela A. Castrillo

Department of Entomology
Cornell University
Ithaca, NY, USA

Jenny S. Cory

Department of Biological Sciences
Simon Fraser University
Burnaby, BC, Canada

Surendra K. Dara

University of California Cooperative
Extension
Division of Agriculture and Natural
Resources
San Luis Obispo, CA, USA

Pauline S. Deschodt

Department of Biological Sciences
Simon Fraser University
Burnaby, BC, Canada

Jørgen Eilenberg

Department of Plant and Environmental
Sciences
University of Copenhagen
Frederiksberg, Denmark

Bret D. Elder

Department of Biological Sciences
Louisiana State University
Baton Rouge, LA, USA

James R. Fuxa

Louisiana State University (Retired)
Cypress, TX, USA

Itamar Glazer

Department of Entomology
ARO, Volcani Centre
Rishon LeZion, Israel

Tarryn A. Goble

Department of Entomology
Cornell University
Ithaca, NY, USA

Ann E. Hajek

Department of Entomology
Cornell University
Ithaca, NY, USA

Ivan Hiltbold

Department of Entomology and Wildlife
Ecology
University of Delaware
Newark, DE, USA

Gernot Hoch

Department of Forest Protection
BFW Austrian Research Centre for
Forests
Vienna, Austria

Trevor A. Jackson

AgResearch Ltd
Lincoln Research Centre
Christchurch, New Zealand

Annette Bruun Jensen

Department of Plant and Environmental
Sciences
University of Copenhagen
Frederiksberg, Denmark

Lawrence A. Lacey

IP Consulting International
Yakima, WA, USA

Edwin E. Lewis

Department of Entomology and
Nematology
University of California – Davis
Davis, CA, USA

Dana Ment

Department of Entomology
ARO, Volcani Centre
Rishon LeZion, Israel

Nicolai V. Meyling

Department of Plant and Environmental
Sciences
University of Copenhagen
Frederiksberg, Denmark

Maureen O'Callaghan

AgResearch Ltd
Lincoln Research Centre
Christchurch, New Zealand

Natalie D. Rivlin

Institute of Marine and Environmental
Technology
University of Maryland Baltimore
County
Baltimore, MD, USA

David I. Shapiro-Ilan

USDA-ARS, SEA
SE Fruit and Tree Nut Research Unit
Byron, GA, USA

Jeffrey D. Shields

Department of Aquatic Health Sciences
Virginia Institute of Marine Science
The College of William & Mary
Gloucester Point, VA, USA

Ikkei Shikano

Department of Entomology and Center
for Chemical Ecology
Pennsylvania State University
University Park, PA, USA

Amanda Shore-Maggio

Institute of Marine and Environmental
Technology
University of Maryland Baltimore
County
Baltimore, MD, USA

List of Contributors

Leellen F. Solter

Illinois Natural History Survey
Prairie Research Institute
University of Illinois
Champaign, IL, USA

Trevor Williams

Instituto de Ecología AC (INECOL)
Xalapa, Veracruz, Mexico

Preface

All have their worth and each contributes to the worth of the others.

J.R.R. Tolkien, *The Silmarillion*

When you try to study something in isolation, you find it hooked to everything else.

John Muir

It becomes necessary in any active scientific discipline to sit back every few years and take stock of the “state of the art.” The time arrives to review recent progress, inspire new ideas, and propose critical and novel lines of research.

In 1963, Yoshinori (“Joe”) Tanada laid the foundation for the current book with his chapter “Epizootiology of Infectious Diseases” in E.A. Steinhaus’ *Insect Pathology: An Advanced Treatise*, a two-volume reference that defined the scope of invertebrate pathology. By 1987, after almost a quarter-century, the time had come for more than just a review of the “state of the art.” Fuxa and Tanada formalized the new scientific discipline in their edited monograph, *Epizootiology of Insect Diseases*, organizing an emerging field of study by establishing its components, definitions, types of studies, and research methods.

Another 30 years have passed, and that time has come again. Much has happened since 1987 – science never stands still. New methods have opened doors unheard of in the 1980s, most notably in molecular biology. Detection and characterization of strands of DNA, RNA, and transposable genetic elements create almost unlimited research opportunities in ecology. New diseases have emerged, such as the mysterious colony collapse disorder of the honeybee, which has raised concern throughout much of the world. Previously characterized diseases have erupted again in devastating epizootics, notably MSX disease (*Haplosporidium nelsoni*) and dermo disease (*Perkinsus marinus*) in populations of oysters. New relationships have opened eyes – who would have thought that microsporidia are highly evolved fungi, not primitive protozoans that evolved before the advent of mitochondria? New concepts have arisen for invertebrate pathogens, contributing to theory in general ecology and host–pathogen coevolution.

Thus, we arrive at this new book. The reader, however, might ask, why *Ecology of Invertebrate Diseases* rather than *Epizootiology of Insect Diseases*? A definition of epizootiology borrowed from the 1987 volume, “the science of causes and forms of the mass phenomena of disease at all levels of intensity in a host population,” allows for study of the total environment, including the host and pathogen populations, even a pathogen’s environment inside its host. Epizootiology in turn is a subset of ecology, which was defined by H.G. Andrewartha as “the scientific study of the distribution and abundance of organisms,” a definition that has evolved to embrace concepts of “population” and “ecosystem.”

Simpler may be better. Epizootiology is the study of animal disease at the population level. It fits well into a broader mold of ecology, as outlined by Tolkien and Muir, if they may be paraphrased, that everything is “connected to everything else” and “contributes to the worth of the others.” Perhaps these two authors were not trying to define ecology, but they might just as well have been.

The editors of this book realized that the “state of the science” has exceeded the scope of the 1987 monograph, thereby creating a multitude of opportunities to discuss new concepts and types of studies, not to mention the myriad of non-insect hosts of infectious disease. Even in a new volume, however, old questions arise, especially, why study ecology of pathogens and their hosts?

First and foremost, parasites are not just dirty little things living a disgusting lifestyle. They are highly evolved organisms – or near-organisms – as intricate and unique as any creature on the planet. They contribute to all, whether by culling the weak or by transporting bits of DNA, in relationships with their hosts ranging from near-benign to something out of a horror film. The reader who delves into this book will see the “worth” in these fascinating little creatures, whether prokaryotic, eukaryotic, or viral.

And, of course, invertebrate pathogens certainly are “hooked to everything” abiotic and biotic, even to humans, a web of life and environment and planet earth. Many such interactions almost defy belief, for that is life.

Science also is called upon to provide tangible benefit. The historical advantages of studying invertebrate diseases remain as important as ever – enhancing disease in pestiferous organisms and preventing disease in invertebrates useful to humans. Pathogens, even viruses, function as parasites with population-level and ecological characteristics, parasites that must be suppressed in populations of beneficial organisms or conserved or enhanced if they are to succeed in pest management. Critics, however, might say that such “germ warfare” against pests is passé, that we now have genetically modified crops, recombinant mosquitoes, and on and on. Perhaps. However, the biopesticide market, which is based on mass production of invertebrate pathogens, continues to grow. Moreover, nothing works in isolation. For example, understanding the dynamics of insect population resistance to disease facilitated management of resistance in widespread use of crops incorporating toxin genes and environmental risk assessment contributed to a safe, first release of a recombinant baculovirus.

Diseases of invertebrate hosts, many of them easy to culture, with their tiny sizes and short generation times, also serve as model systems giving insight into disease ecology in higher organisms – for example, contributions of pathogen reproductive rate, transmission, and virulence to epizootics.

So much for the “What?” and the “Why?” of this book – how about the “Who?” Many decisions were made in compiling this monograph, not the least of which is, where does one stop if everything is “hooked to everything else”? Researchers will find themselves fortunate in this volume’s scientific writers, fortunate because they will recognize the names of the editors and authors, all outstanding pathologists, ecologists, or epizootiologists. Readers will appreciate as they peruse this book that, much like Tolkien’s world, every author has indeed “contributed to the worth of the others.”

James R. Fuxa
Louisiana State University (Retired)

Section I

Introduction

General Concepts in the Ecology of Invertebrate Diseases

Ann E. Hajek¹ and David I. Shapiro-Ilan²

¹ Department of Entomology, Cornell University, Ithaca, NY, USA

² USDA-ARS, SEA, SE Fruit and Tree Nut Research Unit, Byron, GA, USA

1.1 Introduction

With the advent of molecular methods, new species of pathogens and parasites are constantly being described, and as these new species are found, we are learning more about the ecology of new invertebrate diseases, as well as diseases known for many years. Parasitism is a specific and common life-history strategy, and understanding the activity of parasites is central to community and population ecology (Bonsall, 2004). Parasitism of invertebrate hosts also has practical sides, because diseases can help to control insects in an environmentally friendly manner, but we also need to understand the ecology of diseases killing beneficial invertebrates, ranging from pollinators to clams and shrimp, in order to protect managed populations.

The ecology of invertebrate diseases is often referred to as the epizootiology of invertebrate diseases; the word **epizootiology** is similar to the term epidemiology but refers specifically to “the science of causes and forms of the mass phenomenon of disease at all levels of intensity in an animal population” (Fuxa and Tanada, 1987). The ecology of animal diseases, with emphasis on vertebrates, has been treated in an edited book on disease ecology (Hudson et al., 2002), followed by books emphasizing community and ecosystem ecology (Collinge and Ray, 2006; Hatcher and Dunn, 2011; Ostfeld et al., 2014).

Disease ecology with an emphasis on invertebrates was first addressed by Steinhaus (1949), specifically in relation to insects, and the treatment of this subject developed depth and breadth with the publication of an edited volume by Fuxa and Tanada (1987). Around this time, Anderson and May (1981, 1982) created models to investigate factors driving the development of disease epizootics, with at least one system involving epizootics caused by a granulovirus in a forest-defoliating lepidopteran (Anderson and May, 1980). Today, studies of the ecology of invertebrate diseases are commonly conducted, often to understand the ecology underpinning control of invertebrate pests by pathogens, or to understand protection from pathogens for invertebrates valued by humans. In addition, ecological studies of invertebrate diseases are used to build theoretical insights into the causes and dynamics of all diseases. With the wealth of knowledge that

has accumulated since the last synthesis on the ecology of invertebrate diseases in 1987, it is high time to pull together information on this subject. We are also broadening the focus of this book to include the ecology of diseases of all invertebrates and not only insects. Therefore, the hosts included in this book range from pest insects like grasshoppers and caterpillars to valued insects like bees, along with marine and soil invertebrates that are important to humans or ecosystems.

In this chapter, we will present and define the basic concepts on which this field of study is built. Concepts that will be defined will be consistent with definitions in the online glossary published by the Society for Invertebrate Pathology (Onstad et al., 2006).

1.1.1 What Is Disease?

There are numerous definitions for disease, but we consider **disease** to be a departure from the state of health or normality. Of course, this creates a very broad characterization, including multitudes of causes. However, this book will focus on **infectious** diseases, which are those diseases caused by living organisms. Invertebrates are also hosts to many **noninfectious diseases**, of which physical and chemical injuries, genetic diseases, and cancers are a few examples. An example of noninfectious disease impacting insects can occur due to exposure to pesticides. Noninfectious diseases are, however, outside of the material being covered in this book. Descriptive treatments of noninfectious diseases of a diversity of invertebrates can be found in Lewbart (2012) and Sparks (1972).

Returning to disease being a departure from health, this can be much more difficult to determine for invertebrates than for higher vertebrates. Diseases of invertebrates that cause subacute effects and which do not kill hosts could very well be regularly occurring at low prevalence but going undetected. Perhaps recent studies demonstrating the diversity of previously undetected and unrecognized pathogens occurring in honey bee (*Apis mellifera*) colonies that do not die (see Chapter 14) indicate that departures from health being caused by a diversity of parasites acting together can be quite common. The most frequent way that invertebrate diseases are recognized is due to the death of hosts, so emphasis in this field has been on acute diseases. However, in recent years, investigations have included the impact of disease on host fitness, both for pathogens causing chronic diseases and for acute pathogens, after infection and before host death.

The living organisms causing infectious diseases are **parasites**, which are organisms that live at a host's expense. This is a very successful life-history strategy as it has been estimated that the majority of species on earth have parasitic lifestyles (Price, 1980; Zimmer, 2000). **Pathogens** are defined as microorganisms capable of producing disease under normal conditions of host resistance and rarely living in close association with a host without producing some level of disease. Simply put, pathogens can be thought of as microscopic parasites. In this book, the main groups of pathogens that will be covered are viruses, bacteria, fungi, nematodes, and protists. Although many nematodes are not microscopic, the genera that constitute a special group – entomopathogenic nematodes – kill their hosts with the aid of symbiotic bacteria; these nematodes have traditionally been included and studied within the discipline of invertebrate pathology, and thus we include them as well. With the great diversity of pathogens and hosts being covered, of course examples will be missed. In addition, this is presently an expanding field of study, as new species of pathogens are constantly being found. One good example of

this is the number of different pathogens that infect honey bees, with numerous examples discovered in recent years. We will focus on representatives from the diversity of pathogens and hosts for which we know the most about disease ecology.

Diseases may be **chronic** or **acute** (or somewhere in between). Chronic diseases are of a long duration, and thus the host is expected to survive a relatively long time before expiring, or to die of other causes before the disease can become fatal. Acute diseases of invertebrates are often of a short duration; host mortality or maximum severity is expected to occur within a relatively short time after infection. Certain pathogen groups, such as entomopathogenic nematodes (Heterorhabditidae and Steinernematidae) and their symbiotic bacteria tend to cause acute disease, whereas others, such as most Microsporidia, tend to cause chronic diseases. Within other pathogen groups, chronic versus acute diseases vary among combinations of pathogens and hosts.

1.1.2 Terminology and Measurements

Accurate use of terminology is critical to effective communication in science. In pathology, a number of terms have been used with variable meanings in the literature (Shapiro-Ilan et al., 2005). The terminology used in this book is supported by a widely accepted glossary by Onstad et al. (2006), which is based on an earlier glossary by Steinhaus and Martignoni (1970). Thus, we refer the reader to Onstad et al. (2006) as a reference for definitions that may not be spelled out in this chapter or other chapters within this book. Nonetheless, some of the more common terms in invertebrate pathology are defined and discussed in this section.

1.1.2.1 Prevalence/Incidence

Prevalence and incidence are examples of terms that have been variably defined in the literature. **Prevalence** refers to the total number or proportion of disease cases in a population at a given time. For example, if a survey of 10 000 pecan weevil (*Curculio caryae*) larvae in a population indicates that 4000 of the individuals are infected with the fungus *Beauveria bassiana*, then the prevalence rate is 40%. In contrast, **incidence** is the number or proportion of new cases of a disease within a population during a specific period of time. For example, in a given week, if 200 *C. caryae* larvae were found to be infected with *B. bassiana* within a population of 10 000 larvae, then the incidence rate would 2% for that week. Both terms are important for quantifying infection and disease levels over space and time. The difference lies in incidence emphasizing only new cases versus prevalence including both new and old cases. Therefore, incidence may be more useful in predicting risk or spread of disease within a certain timeframe, while prevalence provides an assessment of the full impact of a disease on a population at a given time.

1.1.2.2 Pathogenicity/Virulence

The terminology of pathogenicity and virulence has also been variably defined in the literature. However, definitions of pathogenicity and virulence in the field of invertebrate pathology have been largely consistent over time, and are in agreement with definitions found in the fields of human pathology and microbiology (Shapiro-Ilan et al., 2005). **Pathogenicity** is the quality or state of being pathogenic: the potential ability to produce disease. **Virulence** is the disease-producing power of an organism: the degree

of pathogenicity within a population or species. Generally, the term pathogenicity is applied to entire populations or species, whereas virulence is usually intended for within-group or within-species comparisons. Yet, both terms can conceivably be applied across any taxonomic level (strain, species, genus, family, etc.), provided that pathogenicity is a qualitative and virulence a comparative measure.

Indeed, for a given pathogen and host, pathogenicity is absolute whereas virulence is variable (e.g., due to strain or environmental effects). Pathogenicity is an all-or-none phenomenon. An organism is either pathogenic to a host or it is not. Pathogenicity can be established based on Koch's postulates (Shapiro-Ilan et al., 2005; Lacey and Solter, 2012). Virulence can be measured using various approaches, such as LD₅₀ or LC₅₀ (dosage or concentration required to cause 50% mortality in the test organisms), LT₅₀ (time required to cause 50% mortality in the test organisms), or comparative mortality or infection rates at a given dosage or concentration (Shapiro-Ilan et al., 2005).

1.1.2.3 Infection/Infectivity

An **infection** is the entry of a microorganism into a susceptible host. The presence of the organism may or may not cause overt disease (obvious pathological effects) in the host. If the infection is not immediately followed by overt disease then it is termed an attenuated infection. There are three types of attenuated infections: latent, carrier state, and microbial persistence. A latent infection is an asymptomatic infection that is in a dormant or stationary phase and is capable of being activated at a later time. In the case of viruses, a latent phase is also called an occult phase (and the virus is an occult virus at that stage). In a carrier state, the infection remains as an inapparent infection (no overt sign of its presence) in the current host but is capable of being transferred to other hosts. Microbial persistence is characterized by the continued presence of a pathogenic microorganism within the host in the absence of overt disease but following a period of overt disease.

Infectivity is the ability to produce infection. Thus, infectivity is a measurable characteristic. Infectivity can be measured by assessing the number of pathogenic units (e.g., spores, virus particles, infective juvenile nematodes, or other propagules) that have entered a host following exposure to a known quantity. Infectivity can be positively affected by the number of infection routes; infection routes include per os (through the mouth), through the cuticle or natural openings in the host, and vertical transmission (see later).

1.1.2.4 Immunity

Immunity is innate or induced resistance to a disease agent. It tends to be species-specific but can also vary within species. Invertebrates have immune systems, although predominantly without memories; they are not known to recover from infections and become resistant to further infections of the same pathogens, as vertebrates often can. Immune responses can be cellular (via hemocytes) or humoral (e.g., via antimicrobial peptides), or they may involve both humoral and cellular reactions (e.g., melanization) (Chapter 4). The effectiveness of immune responses may depend on strain or population variation, age and fitness, and stress factors such as environmental conditions or diet. For example, feeding an improper diet resulted in higher percentages of *Drosophila* being killed by a bacterium in their hemolymph (Galac and Lazzaro, 2011; Howick and Lazzaro, 2014). See Chapter 4 for a more in-depth discussion of immunity.