

Contents

Preface xv

Acknowledgments xvii

SECTION 1 Philosophy and Approach to Diagnostic Parasitology 1

Why Perform This Type of Testing? 2

Travel 2

Control Issues 2

Epidemiologic Considerations 2

Compromised Patients 2

Approach to Therapy 2

Who Should Perform Diagnostic Parasitology Testing? 3

Laboratory Personnel 3

Nonlaboratory Personnel 3

Where Should Diagnostic Parasitology Testing Be Performed? 3

Inpatient Setting 3

Outpatient or Referral Setting 4

Decentralized Setting 4

Physician Office Laboratories 4

Over-the-Counter (Home Care) Testing 4

Field Sites 4

What Factors Should Precipitate Testing? 5

Travel History 5

Immune Status of the Patient 5

Clinical Symptoms 5

Documented Previous Infection 5

Contact with Infected Individuals 5

Screening	6
Occupational Screening	6
Therapeutic Failure	6
What Testing Should Be Performed?	6
Screening Tests	6
Routine Methods	7
Special Testing	7
Other (Nonmicrobiological) Testing	7
What Factors Should Be Considered When Developing Test Menus?	8
Physical Plant	8
Client Base	8
Customer Requirements and Perceived Levels of Service	8
Personnel Availability and Level of Expertise	8
Equipment	8
Budget	9
 SECTION 2 Parasite Classification and Relevant Body Sites	 11
Protozoa (Intestinal)	13
Amebae	13
Flagellates	13
Ciliates	13
Coccidia	13
Microsporidia	13
Protozoa (Other Body Sites)	14
Amebae	14
Flagellates	14
Coccidia	14
Microsporidia	14
Protozoa (Blood and Tissue)	14
Sporozoa	14
Flagellates	15
Nematodes (Intestinal)	15
Nematodes (Tissue)	15
Nematodes (Blood and Tissue)	16
Cestodes (Intestinal)	16
Cestodes (Tissue)	16
Trematodes (Intestinal)	16
Trematodes (Liver and Lung)	17
Trematodes (Blood)	17
Pentastomids	17
Acanthocephala	17
Table 2.1 Classification of Human Parasites	19

Table 2.2 Cosmopolitan Distribution of Common Parasitic Infections 21

Table 2.3 Body Sites and Possible Parasites Recovered 22

SECTION 3 Collection Options 23

Safety 24

Collection of Fresh Stool Specimens 24

Collection Method 24

Number of Specimens 25

Collection Times 25

Specimen Type, Stability, and Preservation 26

Preservation of Stool Specimens 26

Formalin 27

Sodium Acetate-Acetic Acid-Formalin (SAF) 28

Schaudinn's Fluid 29

Polyvinyl Alcohol (PVA) 29

Modified PVA 30

Single-Vial Collection Systems 31

Quality Control for Preservatives 32

Procedure Notes 32

Procedure Limitations 33

Collection of Blood 33

Collection of Specimens from Other Body Sites 34

Table 3.1 Fecal Specimens for Parasites: Options for Collection and Processing 36

Table 3.2 Preservatives and Procedures Commonly Used in Diagnostic Parasitology (Stool Specimens) 38

Table 3.3 Body Sites and Specimen Collection 39

SECTION 4 Specimen Test Options: Routine Diagnostic Methods and Body Sites 41

O&P Examination of Stool Specimens 42

Other Diagnostic Methods for Stool Specimens 43

Culture of Larval-Stage Nematodes 43

Estimation of Worm Burdens through Egg Counts 44

Hatching Test for Schistosome Eggs 44

Screening Stool Samples for Recovery of a Tapeworm Scolex 44

Testing of Other Intestinal Tract Specimens 45

Examination for Pinworm 45

Sigmoidoscopy Material 45

Duodenal Drainage 46

Duodenal Capsule Technique (Entero-Test) 47

Urogenital Tract Specimens 47

Sputum 48

Aspirates	48
Biopsy Specimens	49
Blood	49
Thin Blood Films	50
Thick Blood Films	50
Blood Staining Methods	51
Buffy Coat Films	51
QBC Microhematocrit Centrifugation Method	51
ParaSight F Test	52
Knott Concentration	52
Membrane Filtration Technique	52
Culture Methods	52
Animal Inoculation and Xenodiagnosis	53
Antigen and Antibody Detection	53
Table 4.1 Body Sites, Specimens, and Test Options	56
Table 4.2 Serologic, Antigen, or Probe Tests Used in the Diagnosis of Parasitic Infections	58
 SECTION 5 Specific Test Procedures and Algorithms	 59
Microscopy	60
CALIBRATION OF THE MICROSCOPE	60
O&P Examination	63
DIRECT WET FECAL SMEAR	63
SEDIMENTATION CONCENTRATION (Formalin-Ethyl Acetate)	70
FLOTATION CONCENTRATION (Zinc Sulfate)	74
PERMANENT STAINED SMEAR	80
TRICHROME STAIN	82
IRON HEMATOXYLIN STAIN (Spencer-Monroe Method)	88
IRON HEMATOXYLIN STAIN (Tompkins-Miller Method)	93
MODIFIED IRON HEMATOXYLIN STAIN (Incorporating Carbol Fuchsin Step)	95
POLYCHROME IV STAIN	97
CHLORAZOL BLACK E STAIN	97
Specialized Stains for Coccidia and Microsporidia	99
KINYOUN'S ACID-FAST STAIN (Cold Method)	99
MODIFIED ZIEHL-NEELSEN ACID-FAST STAIN (Hot Method)	102
MODIFIED TRICHROME STAIN FOR MICROSPORIDIA (Weber, Green)	107
MODIFIED TRICHROME STAIN FOR MICROSPORIDIA (Ryan, Blue)	109
MODIFIED TRICHROME STAIN FOR MICROSPORIDIA (Kokoskin, Hot Method)	113

Larval Nematode Culture 115
HARADA-MORI FILTER PAPER STRIP CULTURE 115
BAERMANN CONCENTRATION 117
AGAR PLATE CULTURE FOR *STRONGYLOIDES STERCORALIS* 120

Other Methods for Gastrointestinal Tract Specimens 123
EXAMINATION FOR PINWORM (Cellulose Tape Preparations) 123
SIGMOIDOSCOPY SPECIMENS (Direct Wet Smear) 125
SIGMOIDOSCOPY SPECIMENS (Permanent Stained Smear) 129
DUODENAL ASPIRATES 131

Methods for Urogenital Tract Specimens 134
DIRECT SALINE MOUNT 134
PERMANENT STAINED SMEAR 137
URINE CONCENTRATION (Centrifugation) 140
URINE CONCENTRATION (Nuclepore Membrane Filter) 142

Preparation of Blood Films 145
THIN BLOOD FILMS 145
THICK BLOOD FILMS 147
COMBINATION THICK-THIN BLOOD FILMS 149
GIEMSA STAIN 151

Blood Concentration 154
BUFFY COAT CONCENTRATION 154
KNOTT CONCENTRATION 156
MEMBRANE FILTRATION CONCENTRATION 158

Algorithm 5.1 Procedure for Processing Fresh Stool for the O&P Examination 162

Algorithm 5.2 Procedure for Processing Liquid Specimens for the O&P Examination 163

Algorithm 5.3 Procedure for Processing Preserved Stool for the O&P Examination 164

Algorithm 5.4 Use of SAF-Preserved Stool for the O&P Examination 165

Algorithm 5.5 Use of Various Fixatives and Their Recommended Stains 166

Algorithm 5.6 Ordering Algorithm for Laboratory Examination for Intestinal Parasites 167

Algorithm 5.7 Procedure for Processing Blood for Examination for Parasites 168

Table 5.1 Body Site, Specimen, and Recommended Stain(s) 169

SECTION 6 Commonly Asked Questions about Diagnostic Parasitology 173

Specimen Collection	174
Intestinal Tract	174
Blood	175
Tissues or Fluids	175
Fixatives	175
Specimen Processing	176
O&P Examination	176
Diagnostic Methods	177
Direct Wet Examinations	177
Concentrations	177
Permanent Stains	178
Blood Smear Stains	179
Organism Identification	180
Protozoa	180
Helminths	180
Blood Parasites	181
Reporting	181
Organism Identification	181
Quantitation	181
Proficiency Testing	182
Wet Preparations	182
Permanent Stained Smears	182
Blood Films	182
 SECTION 7 Parasite Identification	 183
Protozoa	184
Amebae (Intestinal)	184
<i>Entamoeba histolytica</i>	184
<i>Entamoeba dispar</i>	186
<i>Entamoeba hartmanni</i>	188
<i>Entamoeba coli</i>	190
<i>Endolimax nana</i>	192
<i>Iodamoeba bütschlii</i>	194
<i>Blastocystis hominis</i>	196
Flagellates (Intestinal)	198
<i>Giardia lamblia</i>	198
<i>Dientamoeba fragilis</i>	200
<i>Chilomastix mesnili</i>	202
<i>Trichomonas hominis</i>	204
<i>Enteromonas hominis</i> , <i>Retortamonas intestinalis</i>	206
Ciliates (Intestinal)	208
<i>Balantidium coli</i>	208
Coccidia (Intestinal)	210
<i>Cryptosporidium parvum</i>	210

<i>Cyclospora cayetanensis</i>	212
<i>Isospora belli</i>	214
Microsporidia (Intestinal)	216
<i>Enterocytozoon bienersi</i>	216
<i>Encephalitozoon intestinalis</i> , <i>Encephalitozoon</i> spp.	218
Sporozoa (Blood and Tissue)	220
<i>Plasmodium vivax</i>	220
<i>Plasmodium falciparum</i>	222
<i>Plasmodium malariae</i>	224
<i>Plasmodium ovale</i>	226
<i>Babesia</i> spp.	228
Flagellates (Blood and Tissue)	230
<i>Leishmania</i> spp.	230
<i>Trypanosoma brucei gambiense</i> , <i>T. brucei rhodesiense</i>	232
<i>Trypanosoma cruzi</i>	234
Flagellates (Other Body Sites)	236
<i>Trichomonas vaginalis</i>	236
Nematodes	238
Intestinal	238
<i>Ascaris lumbricoides</i>	238
<i>Trichuris trichiura</i>	240
<i>Necator americanus</i> , <i>Ancylostoma duodenale</i> (Hookworms)	242
<i>Strongyloides stercoralis</i>	244
<i>Enterobius vermicularis</i>	246
Tissue	248
<i>Trichinella spiralis</i>	248
Blood and Tissue	250
Filarial Worms	250
Cestodes	252
Intestinal	252
<i>Taenia saginata</i>	252
<i>Taenia solium</i>	254
<i>Diphyllobothrium latum</i>	256
<i>Hymenolepis nana</i>	258
<i>Hymenolepis diminuta</i>	260
<i>Dipylidium caninum</i>	262
Tissue	264
<i>Echinococcus granulosus</i>	264
Trematodes	266
Intestinal	266
<i>Fasciolopsis buski</i>	266
Liver and Lungs	268
<i>Paragonimus westermani</i> , <i>Paragonimus mexicanus</i>	268
<i>Fasciola hepatica</i>	270

Clonorchis sinensis 272

Blood 274

Schistosoma spp. (*S. mansoni*, *S. haematobium*, *S. japonicum*,
S. mekongi) 274

Photo/Illustration Credits (Section 7) 276

SECTION 8 Identification Aids 277

Tables 8.1 to 8.35 278–327

Identification Keys 8.1 to 8.4 329–332

Life Cycles 333–334

Figures 8.1 to 8.6 335–341

Index 343