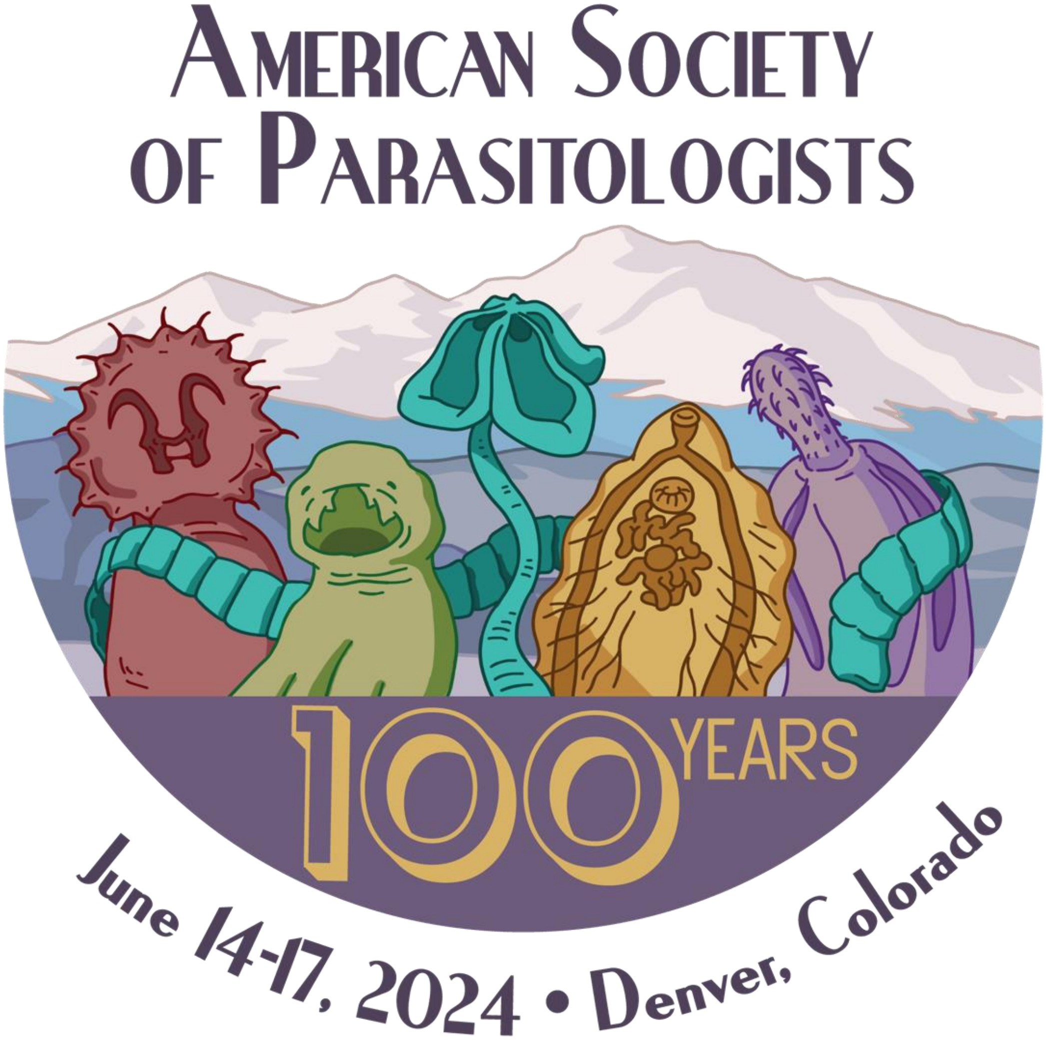
The 99th Annual Meeting
Celebrating 100 years since the inception of the American Society of Parasitologists



Program and Abstracts

Thanks to Everyone Who Helped Make this Meeting Possible …

The American Society of Parasitologists gratefully acknowledges the following for their support, sponsorship, and hard work in putting together this year's annual meeting.

ASP Local Organizing Committee

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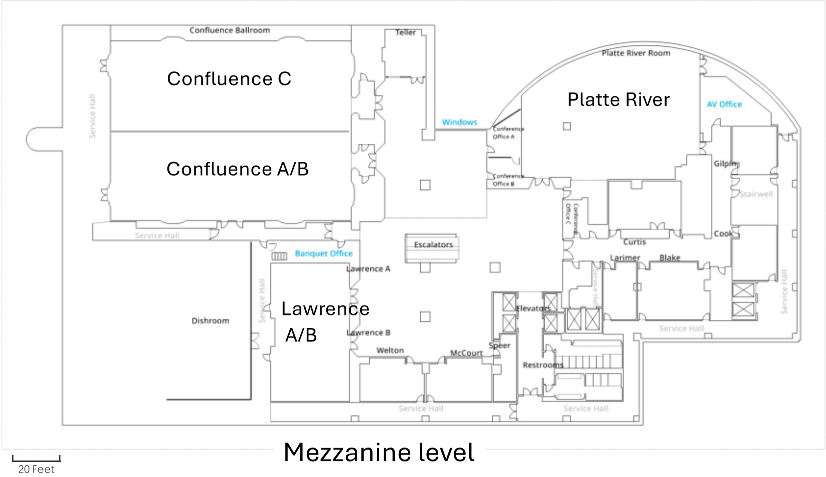
Welcome

We would like to welcome you to the 99th annual meeting of the American Society of Parasitologists (ASP).

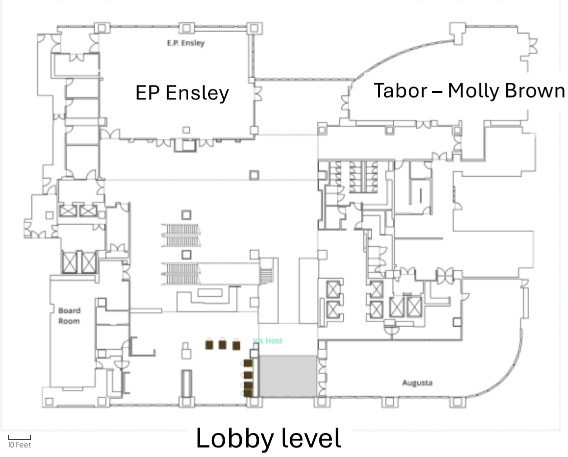
The ASP is a diverse group of approximately 500 scientists from industry, government, and academia who are interested in the study and teaching of parasitology. Founded in 1924, ASP members have contributed not only to the development of parasitology as a discipline, but also to primary research in systematics, medicine, molecular biology, immunology, physiology, ecology, biochemistry, behavior, and more.

Maria G. Castillo and Judith E. Humphries, Scientific Program Officers

Map of meeting rooms



Mezzanine level



Lobby level

American Society of Parasitologist's Discrimination Policy

Statement of Policy: In accordance with the bylaws of the American Society of Parasitologists (ASP), the Society will afford an environment free from discrimination, harassment, and retaliation. The ASP will not tolerate actions, statements, or contacts that discourage the free expression and exchange of scientific ideas. This includes unequal treatment or harassment of any person based on their age, gender, gender identity or expression, marital status, sexual orientation, race, color, national or ethnic origin, religious identifications, beliefs or practices, disabilities, veteran status, or any other reasons or expressions that are unrelated to their scientific merit. Harassment, sexual or otherwise, shall be considered as a form of misconduct and violators will be subject to disciplinary actions, including expulsion from a society function or from the society itself.

Definition of Sexual Harassment: Sexual harassment refers to unwelcome sexual advances, requests for sexual favors, and other verbal or physical conduct of a sexual nature. Sexual harassment does not refer to occasional compliments of a socially acceptable nature. It refers to behavior that is not welcome, is personally offensive, debilitates morale, and therefore, interferes with a collegial atmosphere. The following are examples of behavior that, when unwelcome, may constitute sexual harassment: sexual flirtations, advances, or propositions; verbal comments or physical actions of a sexual nature; sexually degrading words used to describe an individual; a display of sexually suggestive objects or pictures; sexually explicit jokes; unnecessary touching. What is perceived as acceptable to one person may be unwelcome by another. Those who have positions of authority or higher rank should be aware that others may be reluctant to outwardly express objections or discomfort regarding unwelcome behavior or language.

Other Types of Harassment: Remarks and behaviors based on other protected characteristics are also unacceptable to the Society. These include stereotyping, slurs, derogatory jokes or statements, and any hostile or intimidating acts.

Policy Scope: This policy applies to all attendees and participants at ASP meetings and functions, including social functions, tours, or off-site activities during the course of meetings and functions, and includes all members, guests, staff, contractors, and exhibitors.

Reporting an Incident: If any individual covered by this policy believes that they have experienced or witnessed harassment or bullying they should contact the society's designated individual [Dr. Sara Brant, sbrant@unm.edu]. No complainant will be required to discuss any incident with a respondent; no respondent will be required to discuss any incident with a complainant. All individuals (complainant or respondent) may bring an accompanying individual of their choice with them for support at any point when they discuss the matter with the society's designated individual, or during any course of an ensuing investigation.

Because allegations of discrimination, harassment and misconduct are sensitive matters with the potential to negatively impact the reputation of individuals, institutions, and/or our Society, confidentiality and discretion throughout the process is expected from all parties involved and is assured from the ASP's designated individual and all involved in the investigation.

Regardless, a complainant may speak in confidence with the society's designated individual without involving an official report, an investigation or a respondent. All complaints that are received will be treated seriously, and will be addressed promptly if that is the wish of a complainant. Any incidents of sexual assault should be immediately reported to the police. Note that many local and regional governments also consider a variety of behaviors to be reportable crimes regardless of the wishes of the complainant, respondent or of the society.

Investigation: Following the official report of an incident, the Society's designated individual, in consultation with ASP Council, will name an impartial investigator; usually an elected officer or Council member, and the respondent will be promptly notified. No one who has a conflict of interest with respect to the complainant or respondent will serve in this role. A complainant will be asked to file a formal written complaint; the respondent will be notified immediately and prior to any discovery procedures. A respondent will be invited to respond to the complaint and allowed to bring evidence. The Council of ASP reserves the right to interview other individuals as witnesses at its own discretion. The investigator is allowed to seek counsel if they are in doubt as to how to proceed.

When the investigation is complete, the findings will be communicated to the elected officers, as well as both to the complainant and respondent. Those officers without a conflict of interest will decide on appropriate disciplinary actions.

Retaliation: The Society will not tolerate any form of retaliation against individuals who report an incident, against those who are subject to a complaint, nor against those who participate in an investigation. Retaliation will be considered a form of discrimination in and of itself and offenders will be subject to disciplinary action, up to and including ejection from the society.

Disciplinary Action: If an individual harasses, retaliates, or knowingly makes a false claim, they will be subject to disciplinary action. These actions might range from a verbal warning to a request to leave the meeting or function without refund of fees and a reporting of the incident to the person’s employer. Should repeated complaints, patterns of inappropriate behavior, or other events emerge, the society’s by-laws permit its Council to exclude and eject members through a process that has no appeal.

Appeal & Questions: Should any person be dissatisfied with the result of an investigation or disciplinary action, they may appeal to the President of the Society, or to the highest-ranking officer without a conflict of interest. Questions concerning the policy can be directed to an ASP officer or the ASP designated individual.

THE AMERICAN SOCIETY OF PARASITOLOGISTS
1924

General Schedule

Day/Times	Activity/Function	Room/Space
Friday June 14		
7:00 am–5:00 pm	Registration	Conference Office
8:00 am–Noon	ASP Council Meeting	Molly Brown & Horace Tabor
2:30 pm–4:35 pm	Special Symposium: Innovative Teaching for Parasitology	Confluence C
2:30 pm–4:30 pm	Ecology & Evolution I	Confluence A/B
2:10 pm–4:30 pm	Special Symposium: 2nd International Nematomorpha Symposium	Lawrence A/B
4:30 pm–4:50 pm	Coffee Break	Mezzanine Foyer
4:50 pm–6:05 pm	Immunology, Vector Biology	Confluence C
4:50 pm–6:20 pm	Genomics & Molecular Biology I	Confluence A/B
4:50 pm–6:05 pm	Biochemistry/Physiology, Chemotherapy/Drug Resistance	Lawrence A/B
6:30 pm–10:00 pm	Welcome Reception (Full dinner)	Mezzanine Foyer
Saturday June 15		
7:00 am–5:00 pm	Registration	Conference Office
8:20 am–10:30 am	ASP President's Symposium	Confluence A/B/C
10:30 am–10:50 am	Coffee Break	Mezzanine Foyer
10:50 am–Noon	President's Address	Confluence A/B/C
Noon–1:30 pm	Editor's Lunch (by invitation)	Molly Brown
1:30 pm–3:15 pm	Host-Parasite Interactions I	Confluence C
1:30 pm–3:15 pm	Life Cycles & Epidemiology, Cell Biology	Confluence A/B
1:30 pm–3:05 pm	Special Symposium: Methodological Frontiers for Characterizing Parasite Communities	Lawrence A/B
2:00 pm–3:30 pm	Auction Set Up	Platte River
3:30 pm–4:00 pm	Auction Preview	Platte River
4:00 pm–6:00 pm	ASP Student Auction (Snacks and Beverages)	Platte River

Sunday June 16

7:00 am–11:00 am	Registration	Conference Office
8:30 am–10:00 am	Genomics & Molecular Biology II, Life cycles & Epidemiology II	Lawrence A/B
8:00 am–10:00 am	Taxonomy, Systematics, & Phylogeny I	Confluence C
7:50 am–10:00 am	Special Symposium: Host-Parasite Interactions in Marine Ecosystems I	Confluence A/B
10:00 am–10:20 am	Coffee Break	Mezzanine Foyer
10:20 am–Noon	Students' Symposium	Confluence C
10:20 am–Noon	Special Symposium: Host-Parasite Interactions in Marine Ecosystems II	Confluence A/B
Noon–1:30 pm	Student's Luncheon & Vortex	Platte River
1:30 pm–3:30 pm	Host-Parasite Interactions II	Confluence C
1:30 pm–3:30 pm	Taxonomy, Systematics, & Phylogeny II	Confluence A/B
1:30 pm–3:30 pm	Ecology & Evolution II	Lawrence A/B
3:30 pm–4:30 pm	Students' Business Meeting	Platte River
4:30 pm–6:30 pm	Poster Session and Palettes & Parasites (Snacks and Beverages)	Molly Brown & Horace Tabor

Monday June 17

8:15 am–10:00 am	Taxonomy, Systematics, & Phylogeny III	Platte River
8:00 am–10:00 am	Host-Parasite Interactions III	Molly Brown & Horace Tabor
8:00 am–10:00 am	Ecology & Evolution III	Lawrence A/B
10:00 am–10:20 am	Coffee Break	Mezzanine Foyer
10:20 am–11:20 am	Eminent Parasitologist Lecture	Confluence A/B/C
1:00 pm–2:00 pm	H.B. Ward Medal Lecture	Confluence A/B/C
2:00 pm–4:30 pm	ASP Awards & Business Meeting*	Confluence A/B/C

*Awards being presented include the Clark P. Read Mentor Award, Ashton Cuckler New Investigator Award, Marc Dresden Student Travel Grants, and Best Student Presentations.

Note: All food indicated in the schedule will be included with regular conference registration. A full dinner will be provided at the welcome reception on June 14th (with regular conference registration). Lunch will be provided at the student vortex lunch on June 16th; all students are invited, and non-students are by invitation only. Coffee breaks will be provided. Drink tickets will be provided along with a cash bar for several evening events. Light snacks will be provided at the auction and poster sessions (Sat and Sun) and these sessions will end on time for attendees to plan their own dinner reservations outside the venue on those evenings, dinner reservations are recommended.

Detailed Schedule

Friday, June 14

7:00 am–5:00 pm	**Registration**	**Conference Office**
8:00 am–Noon	**ASP Council Meeting**	**Molly Brown & Horace Tabor**
2:30–4:35 pm	**Special Symposium: Innovative Teaching for Parasitology**	**Confluence C**

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Valerie McKenzie**, University of Colorado, Boulder, CO
Elliot Zieman, Eastern Illinois University, Charleston, IL

2:30 Opening Remarks

2:40 (1) **Christina Anaya.** INTEGRATING ACTIVE LEARNING STRATEGIES INTO PARASITOLOGY COURSES TO PROMOTE STUDENT-CENTERED LEARNING.

2:55 (2) **Sarah A Orlofske.** FLIPPING OUT: OPPORTUNITIES AND CHALLENGES FOR PARASITOLOGY IN A “FLIPPED” CLASSROOM.

3:10 (3) **Ana E Garcia Vedrenne.** INCORPORATING TECHNOLOGY SUPPORTED, TEAM-BASED LEARNING INTO A PARASITOLOGY COURSE.

3:25 (4) † **Joseph T Cornelius**, Joanna J Cielocha. ASSESSMENT OF STUDENT LEARNING OUTCOMES AND PERCEPTIONS OF ACTIVE LEARNING IN AN UNDERGRADUATE PARASITOLOGY COURSE.

3:40 (5) **John F Shea**. USING THE IMAGINATION TO TEACH PARASITOLOGY WITH CASE STUDIES AND CREATIVITY.

3:55 (6) † **Kevin Garcia**, Ana Elisa Garcia-Vedrenne. ENHANCING LEARNING OUTCOMES: SPECIFICATIONS GRADING IN PARASITOLOGY.

4:10 (7) **Hannah Bialic**. DESIGNING EFFECTIVE PUBLIC ENGAGEMENT IN PARASITOLOGY.

4:25 Discussion and Concluding Remarks

2:30–4:30 pm Ecology & Evolution I Confluence A/B

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding:	**Florian Reyda**, State University of New York at Oneonta, Oneonta, NY
2:30 (8)	**Daniela de Angeli Dutra**, Robert Poulin. NETWORK SPECIFICITY DECREASES COMMUNITY STABILITY AND COMPETITION AMONG AVIAN HAEMOSPORIDIAN PARASITES AND THEIR HOSTS.
2:45 (9)	**Sophie EM Elliott**, Dana M Calhoun, Pieter TJ Johnson. LEFT HIGH AND DRY: THE INTERACTIONS BETWEEN TREMATODES AND POND DRYING EVENTS IN BOULDER, COLORADO.
3:00 (10)	**Hannah G Whitcomb**, Florian Reyda. PAST VS. PRESENT: A SURVEY OF THE FISH PARASITES OF THE TRIBUTARIES OF ONEIDA LAKE, NEW YORK.
3:15 (11) †	**Katerina D Sawickij**, Christopher Blanar, J Matthew Hoch, David Kerstetter, Florian Reyda. PARASITES OF THE EASTERN EVERGLADES, FLORIDA FISHES: BANGHAM 1940 REVISITED.
3:30 (12) †	**Maria R Bollinger**, Taylor M Fiedor, Kyle D Gustafson. THE COCCIDIA SPECIES *PFEIFFERINELLA ELLIPSOIDES* EXHIBITS SPATIAL AND SPECIES-SPECIFIC VARIATION IN PREVALENCE AMONG FRESHWATER SNAIL HOSTS.
3:45 (13) †	**Brooke A McPhail**, Patrick C Hanington. HOW DOES DIGENEAN TREMATODE HOST SPECIFICITY AFFECT THE BIODIVERSITY-DISEASE RELATIONSHIP?
4:00 (14)	**Samuel D Ramsey**, Daniel Sonenshine, Steven C Cook, Connor Gulbronson, Jay Evans, Justin Hein, Mikayla Henry, Chris Borke, Madison Sankovitz, Ron Ochoa, Dennis vanEngelsdorp. PARASITIC VARROA MITES EMPLOY KLEPTOPARASITISM ON THE MOLECULAR LEVEL TO ACCELERATE REPRODUCTION.
4:15 (15) †	**Kate P Harder**, Sarah England, Kristina Naden. CANINE HOOKWORM PRESENCE IN TONGATAPU, TONGA.

2:10–4:30 pm Special Symposium: Lawrence A/B

2nd International Nematomorpha Symposium

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding:	**Matt Bolek**, Oklahoma State University, Stillwater, OK **Ben Hanelt**, University of New Mexico, Albuquerque, NM **Christina Anaya**, Florida Gulf Coast University, Fort Myers, FL
2:10	Opening Remarks

2:20 (16) **Ming-Chung Chiu**. REGULATION OF SEXUAL DIFFERENTIATION IN MANTIDS: A NOVEL PERSPECTIVE ON THE IMPACT OF HAIRWORM ON HOST DEVELOPMENT.

2:35 (17) † **Louise M Coates**, Mara Carey-Wood, Chris Vennard, Dominika Lastik, Kieran Reynolds, Jean-François Doherty, Eddy J Dowle, Vicky Hunt. BEHAVIOURAL AND PROTEOMIC ANALYSIS OF THE ROLE OF DEHYDRATION IN CRICKETS, *ACHETA DOMESTICUS*, INFECTED WITH THE NEMATOMORPH *PARAGORDIUS VARIUS.*

2:50 (18) **Jean-François Doherty**, Bingzong Liu, Ben Hanelt, Eddy J Dowle, Neil J Gemmell, Robert Poulin, Leonard J Foster, Benjamin J Matthews. EXPLORING THE SENSORY LANDSCAPE OF HAIRWORM-INFECTED CRICKETS.

3:05 (19) **Ben Hanelt**. BACTERIAL RESISTANCE AND LONGEVITY OF CRICKETS INFECTED WITH GORDIAN WORMS (NEMATOMORPHA).

3:20 (20) **John F Shea**, Emily Klawiter, Nicholas FitzGerald. HAIRWORM ABUNDANCE AND PREVALENCE AS AN INDICATOR OF ECOSYSTEM HEALTH.

3:35 (21) **Christina Anaya**, Lambda Leonov, Guadalupe Rodriguez. THE HAIRWORM AND THE CRICKET: A STORY OF LIFE AND DEATH.

3:50 (22) **Matthew G Bolek**, Christina Anaya, Ben Hanelt. LIFE HISTORY DIVERSITY OF THE FRESHWATER GORDIIDS (PHYLUM: NEMATOMORPHA).

4:05 (23) **Andreas Schmidt-Rhaesa**. BIODIVERSITY OF HORSEHAIR WORMS (NEMATOMORPHA): ADVANCES AND CHALLENGES.

4:20 Discussion and Concluding Remarks

4:30–4:50 pm Coffee Break Mezzanine Foyer

4:50–6:05 pm Immunology, Vector Biology Confluence C

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Nicolas Wheeler,** University of Wisconsin-Eau Claire, Eau Claire, WI

4:50 (24) † **Allison Segard**, William Tolbert, Christopher Broder, Marzena Pazgier, Edward Mitre. BMA-LAD-2 AS A POSSIBLE VACCINE TARGET AGAINST *BRUGIA MALAYI*.

5:05 (25) † **Emily R Hannon**, Youssef Idaghdour, Arlene E Dent, Paula Embury, Sidney Ogolla, David Midem, Scott M Williams, James W Kazura. PERIPHERALLY CIRCULATING IMMUNE CELL PROFILES IN AFRICAN CHILDREN INDICATE A HETEROGENEOUS IMMUNE RESPONSE AGAINST *PLASMODIUM FALCIPARUM* MALARIA.

5:20 (26) **Gul Ahmad**. SCHISTOSOMIASIS: PRINCIPAL REQUIREMENTS FOR THE DEVELOPMENT OF A MOLECULARLY DEFINED VACCINE.

5:35 (27) † **Christina R Bowhay**, Patrick C Hanington. TOLL-LIKE RECEPTORS IN SNAIL HEMOCYTES AND BEYOND.

5:50 (28) † **Cole J Meier**, Shabbir Ahmed, Julián F Hillyer. THE EXTRACELLULAR MATRIX PROTEINS PERICARDIN AND LONELY HEART MEDIATE PERIOSTIAL HEMOCYTE AGGREGATION IN THE MOSQUITO *ANOPHELES GAMBIAE*.

4:50–6:20 pm Genomics & Molecular Biology I Confluence A/B

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Egie E Enabulele,** Texas Biomedical Research Institute, San Antonio, TX

4:50 (29) † **Ijeoma C Okoye**, Ian M Lamb, Swati Dass, Joanne M Morrisey, Michael W Mather, Akhil B Vaidya. *PF*ATAD3 IS A PARASITE-ENCODED PROTEIN CRITICAL FOR MITOCHONDRIAL BIOGENESIS IN *PLASMODIUM FALCIPARUM.*

5:05 (30) † **Elizabeth G Zeldenrust**, Alexandre N Léveillé, John R Barta. EXTREME DIVERSITY OF MITOCHONDRIAL GENOME ORGANIZATION WITHIN THE COCCIDIAN SUBORDER ADELEORINA.

5:20 (31) † **Natasha Slawnych**, Perryn Kruth, Larsen Iorgovitz, Alex Leveille, John Barta. MITOCHONDRIAL GENOMES OF COCCIDIA WITH MORPHOLOGICALLY DISTINCT SPOROCYST EXCYSTATION STRUCTURES.

5:35 (32) **Jenny G Maloney**, Monica Santin. METHODS FOR GENERATING GENOMES OF PROTOZOAN PARASITES WITH FOOD SAFETY IMPORTANCE DIRECTLY FROM FECAL ISOLATES.

5:50 (33) **Monica Santin**, Jenny G Maloney. APPLICATION OF NEXT GENERATION AMPLICON SEQUENCING TO CHARACTERIZE GENETIC DIVERSITY OF PROTOZOAN PARASITES OF FOOD SAFETY IMPORTANCE.

6:05 (34) **Kaylee S Herzog**, Lisa M Harrison, Michael E Wilson, Michael Cappello, Joseph R Fauver. CHARACTERIZING GENETIC DIVERSITY AND POPULATION STRUCTURE OF HUMAN HOOKWORMS USING WHOLE GENOME DATA FROM ACCESSIBLE SAMPLE TYPES.

4:50–6:05 pm Biochemistry/Physiology, Chemotherapy/Drug Resistance Lawrence A/B

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Tom Pennance,** Western University of Health Sciences, Lebanon, OR

4:50 (35) Shannon Kitchen, Elise L McKean, **John M Hawdon**. LOSS OF PHENOTYPIC TRAITS IN HOOKWORMS ASSOCIATED WITH RECOVERY FROM CRYOPRESERVATION.

5:05 (36) **Catherine A Jackson**, Elise L McKean, Andrea Langeland, John M Hawdon. INVESTIGATING THE ANTHELMINTIC POTENTIAL OF *MOMORDICA CHARANTIA* AGAINST HOOKWORM INFECTIONS.

5:20 (37) **Benjamin M Rosenthal**, Matthew Tucker, Christina Yeager, Alan Gutierrez, Manan Sharma, Kalmia Kniel, Mark Jenkins. DEVELOPMENT OF *EIMERIA* SURROGATES TO ADVANCE CONTROL OF HUMAN *CYCLOSPORA* INFECTIONS.

5:35 (38) † **Philip Yeboah**, Hafiz Abdullah, Kaylin Chasser, Audrey Duff, Alejandro Relling, Lisa Bielke. ANTICOCCIDIAL SENSITIVITY AND RESISTANCE PROFILES FROM COMMERCIAL TURKEY FLOCKS IN THE UNITED STATES.

5:50 (39) **Elise L McKean**, John M Hawdon. PHENOTYPIC DIFFERENCES IN FEEDING BEHAVIOR OF MULTI-ANTHELMINTIC DRUG RESISTANT *ANCYLOSTOMA CANINUM* LARVAE SUGGEST MUTATED NICOTINIC ACETYLCHOLINE RECEPTOR.

6:30–10:00 pm Welcome Reception Mezzanine Foyer

(Full Dinner)

Saturday, June 15

7:00 am–5:00 pm Registration Conference Office

8:20–10:30 am ASP President's Symposium Confluence A/B/C

Presiding: **Reginald Blaylock,** University of Southern Mississippi**,** Ocean Springs, MS

Theme: "Parasitology: Past, Present, and Future"

8:20 am Opening Remarks

8:30 (40) **Sascha L Hallett,** Stephen D Atkinson. DEMYSTIFYING MYXOZOANS.

9:00 (41) **W Evan Secor**. HUMAN SCHISTOSOMIASIS: FROM HIEROGLYPHS TO ARTIFICIAL INTELLIGENCE.

9:30 (42) **Emily J Jenkins,** Emilie Bouchard, Geraldine Gouin, Adrian Hernandez-Ortiz. A ONE HEALTH APPROACH TO *TOXOPLASMA GONDII* IN THE ARCTIC.

10:00 (43) **Kevin Lafferty**. ARE PARASITOLOGISTS OBSOLETE (AGAIN)?

10:30–10:50 am Coffee Break Mezzanine Foyer

10:50 am–Noon President's Address Confluence A/B/C

Presiding: **Herman Eure**, Wake Forest University, Winston-Salem, NC

10:50 am **Herman Eure**. Introduction of Reginald Blaylock.

11:00 am **Reginald Blaylock**.

"It's Not Always About The Parasites"

Noon–1:30 pm Editor's Lunch (by invitation only) Molly Brown

1:30–3:15 pm Host-Parasite Interactions I Confluence C

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Ashleigh B Smythe,** Virginia Military Institute, Lexington, VA

1:30 (44) † **Lisa R MacLeod**, Lien T Luong. ECOLOGY OF FEAR: ONTOGENY-MEDIATED NON-CONSUMPTIVE EFFECTS.

1:45 (45) † **Andrea Langeland**, Elise L McKean, Catherine A Jackson, Damien M O'Halloran, John M Hawdon. HOST SPECIFICITY IN HOOKWORMS: SPECIES-SPECIFIC AND SEX-DEPENDENT IMMUNE MECHANISMS MEDIATE HOST PERMISSIVENESS.

2:00 (46) † **Nicholas R Lee**. ENDOSYMBIOTIC *WOLBACHIA PIPIENTIS* IN ONCHOCERCID NEMATODES: THE POTENTIAL ROLE OF BACTERIOPHAGE DNA IN THE BIOLOGY OF THIS HOST-PARASITE SYSTEM.

2:15 (47) † **Caleb I Larsen**, Keri A Mitchell, Abby S Bernier, Sara B Weinstein. STABILITY OF THE *HELIGMOSOMOIDES POLYGYRUS* MICROBIOME FOLLOWING HOST ANTIBIOTIC EXPOSURE.

2:30 (48) † **Hayden A Krause**, Sascha Hallett, Sarah Bjork, Mark Hereford, Ted Wise. AN ASSESSMENT OF PATHOGEN RISK TO JUVENILE SPRING-RUN CHINOOK SALMON (*ONCORHYNCHUS TSHAWYTSCHA*) IN THE UPPER KLAMATH BASIN.

2:45 (49) † **Abiodun Abosede Egbunu**. EVALUATING THE EXPOSURE OF PRIMARY SCHOOL PUPILS TO SOIL TRANSMITTED ASCARIASIS THROUGH WASTEWATER AND DISPOSAL IN LOKOJA KOGI STATE.

3:00 (50) **Asma Abdul Latif**, Tooba Arooj, Najia Al Arifa. INCIDENCE OF INTESTINAL PARASITIC INFECTIONS IN SCHOOL CHILDREN OF FAISALABAD, PAKISTAN.

1:30–3:15 pm Life Cycles & Epidemiology I, Cell Biology Confluence A/B

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Jillian Detwiler,** University of Manitoba, Winnipeg, Manitoba

1:30 (51) † **Anna M Poslednik**, Ryan B Carnegie. VISUALIZATION OF ENVIRONMENTAL CELL FORMS TO BETTER UNDERSTAND THE ECOLOGY AND EVOLUTION OF AN IMPORTANT MARINE PATHOGEN.

1:45 (52) † **Nilanjana Das**, Sarah J Bjork, Stephen D Atkinson, Michael L Kent, Aimee N Reed, Sascha L Hallett. THE EMERGENCE OF *TETRACAPSULOIDES BRYOSALMONAE* IN AN OREGON SALMONID HATCHERY.

2:00 (53) Cameron R Hodinka, **Jillian T Detwiler**. DEVELOPING METHODS TO MAXIMIZE LAB INFECTION SUCCESS OF *ECHINOSTOMA TRIVOLVIS* LINEAGE C IN GASTROPOD FIRST INTERMEDIATE HOSTS.

2:15 (54) **Benjamin M Rosenthal**, Peter C Thompson, Valsin Fournet, Jitender P Dubey. *TRICHINELLA* CONTROL: A SINGULAR PUBLIC HEALTH ACHIEVEMENT.

2:30 (55) **Taylor Clarkson**, Dickson Osabutey, Jeffery Sumboh, Amanda Lamptey, Francis Appiah-Twum, Daniel Obumang, Christopher Dorcoo, Joseph Quartey, Joseph Fauver, Michael Wilson, Michael Cappello. ONE YEAR FOLLOW UP OF A CROSS-SECTIONAL STUDY OF HOOKWORM INFECTION AND RESPONSE TO DEWORMING IN BEPOSO, GHANA.

2:45 (56) Jose Maravi-Jaime, **David Castaneda-Carpio**, Segundo Del Aguila, Renzo Gutierrez-Loli, Cristina Guerra-Giraldez. INSIGHTS ON THE TRANSCRIPTOME ANALYSIS OF THE *TAENIA SOLIUM* DEVELOPMENT FROM METACESTODE TO JUVENILE – ADULT.

3:00 (57) **Uche Chijioke Ngenegbo**, Obiora Osegboka Ikpeze. MALARIA VECTOR CONTROL WITH INDOOR RESIDUAL SPRAYING IN STUDENTS' HOSTELS AT AWKA – NIGERIA.

1:30–3:05 pm Special Symposium: Lawrence A/B
Methodological Frontiers for Characterizing Parasite Communities

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Dan Preston**, Colorado State University, Fort Collins, CO
Georgia Titcomb, Colorado State University, Fort Collins, CO

1:30 Opening Remarks

1:40 (58) **Daniel L Preston**, Landon P Falke, Sara V Brant. SHIFTS IN TREMATODE PARASITE COMMUNITY STRUCTURE FROM HEADWATERS TO A MAINSTEM RIVER IN THE WILLAMETTE RIVER BASIN, OREGON.

1:55 (59) † **Timothy L Odom**, Andrew W Park, Joseph A Cook, Jonathan L Dunnum, Schuyler W Liphardt, Katrina M Derieg, Stephen E Greiman. SEASONAL SUCCESSION OF CESTODE METACOMMUNITIES IN TWO NORTH AMERICAN *SOREX* SHREWS.

2:10 (60) **Jasmine N Childress**. GREATER THAN THE SUM OF ITS PARTS: INTEGRATING DNA METABARCODING AND CLASSICAL METHODS IN PARASITOLOGY TO BETTER UNDERSTAND PARASITE COMMUNITY DYNAMICS.

2:25 (61) † **Leah A Owens**, Sagan Friant, Tony L Goldberg. CHARACTERIZING BIAS, ERROR, AND QUANTITATIVE POTENTIAL IN 18S METABARCODING OF PARASITE ASSEMBLAGES.

2:40 (62) **Elizabeth M Warburton**, Sarah A Budischak, Anna E Jolles, Vanessa O Ezenwa**.** HOST TRAITS, ABIOTIC VARIATION, AND COMMUNITY SIMILARITY: PARASITE Β-DIVERSITY ACROSS LIFE STAGES AND SCALES.

2:55 Discussion and Concluding Remarks

2:00–3:30 pm	**Auction Set Up**	**Platte River**
3:30–4:00 pm	**Auction Preview**	**Platte River**
4:00–6:00 pm	**33rd Annual ASP Student Auction** **(Snacks and Beverages)**	**Platte River**

Sunday, June 16

7:00–11:00 am	**Registration**	**Conference Office**
8:30–10:00 am	**Genomics & Molecular Biology II, Life Cycles & Epidemiology II**	**Lawrence A/B**

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Sascha Hallett**, Oregon State University, Corvallis, OR
Nilanjana Das, Oregon State University, Corvallis, OR

8:30 (63) **Aditya Gupta**, Larissa S de Araujo, Asis Khan, Benjamin M Rosenthal, Jitender P Dubey. MOLECULAR CHARACTERIZATION OF A *SARCOCYSTIS BOVIFELIS* -LIKE SARCOCYSTS IN AMERICAN BEEF**.**

8:45 (64) **Larissa S de Araujo**, Aditya Gupta, Asis Khan, Benjamin M Rosenthal, Jitender P Dubey. HIGH, BUT VARIABLE PREVALENCE OF *SARCOCYSTIS* INFECTIONS IN THE AMERICAN BISON (*BISON BISON*) BEEF DESTINED FOR HUMAN CONSUMPTION.

9:00 (65) Corinne L Conlon, **Christopher M Whipps**. SURVEILLANCE OF THE ZOONOTIC PARASITE *ECHINOCOCCUS MULTILOCULARIS* IN NEW YORK STATE SUGGESTS MULTIPLE INTRODUCTIONS OF THE EUROPEAN VARIANT IN WILD CANID POPULATIONS.

9:15 (66) **Peter Halvarsson**. SPECIES DIVERSITY AND HOST SPECIFICITY AT THE WILDLIFE-FARM ANIMAL INTERFACE

9:30 (67) **Egie E Enabulele**, Neal R Platt, Bonnie L Webster, Aidan M Emery, Grace O Ajakaye, Mahmud Umar Ali, Ebube C Amaechi, Tolulope E Atalabi, Timothy Auta, Oluwaseun B Awosolu, Adamu G Dagona, Chika E Ejikeugwu, Christopher Igbeneghu, Victor S Njom, Mariam Onwude-Agbugui, Nkeiruka M Orji, Funso P Oyinloye, Christopher Pam, Uchenna I Ugah, Timothy J Anderson. HOW DIAGNOSTIC ARE RIBOSOMAL ITS MARKERS FOR SCHISTOSOME SPECIES IDENTIFICATION AND CHARACTERIZATION OF PUTATIVE HYBRIDS?

9:45 (68) **Joseph R Fauver**, Kaylee S Herzog. USING POPULATION GENOMICS AS A TOOL TO EVALUATE GLOBAL HELMINTH CONTROL PROGRAMS.

8:00–10:00 am Taxonomy, Systematics, & Phylogeny I Confluence C

Time (Abstract No.
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Tyler Achatz**, Middle Georgia State University, Macon, GA
Zoe Von Holten, Middle Georgia State University, Macon, GA

8:00 (69) **Philippe Vieira Alves**, Reinaldo J da Silva, Daniel Janies, Willian Taylor, April Harris, Gari New, Denis Jacob Machado. MITOGENOME ORGANIZATION, DIVERSITY, AND EVOLUTIONARY RELATIONSHIPS OF PROTEOCEPHALIDEAN TAPEWORMS (CESTODA, ONCHOPROTEOCEPHALIDEA) UNVEILED BY A GENOME SKIMMING APPROACH.

8:15 (70) **Kirsten Jensen**, Janine N Caira, Veronica M Bueno. NEW DATA, NEW INSIGHTS: A REVISED PHYLOGENY OF THE LECANICEPHALIDEA.

8:30 (71) Philippe Vieira Alves, Reinaldo J da Silva, Alain de Chambrier, José L Luque, Anastasiia Duchenko, Daniel Janies, **Denis Jacob Machado**. MACHINE LEARNING MODELS ACCURATELY PREDICT CLADES OF PROTEOCEPHALIDEAN TAPEWORMS (CESTODA, ONCHOPROTEOCEPHALIDEA) BASED ON HOST AND BIOGEOGRAPHICAL DATA.

8:45 (72) **Kevin Lievano**. MORPHOLOGICAL AND MOLECULAR ANALYSIS OF *MATHEVOTAENIA* (CESTODA: ANOPLOCEPHALIDAE) FROM THE GIANT ANTEATER, *MYRMECOPHAGA TRIDACTYLA* (PILOSA: MYRMECOPHAGIDAE) IN COLOMBIA.

9:00 (73) **Tim R Ruhnke**, Kirsten Jensen, Janine N Caira, Veronica M Bueno. PROGRESS IN THE PHYLOGENY AND CLASSIFICATION OF THE RHINEBOTHRIIDEA.

9:15 (74) **Spencer Galen**. HOW MANY SPECIES OF AVIAN HAEMOSPORIDIAN ARE THERE?

9:30 (75) **Richard E Clopton**, Callie States, Debra T Clopton. *GREGARINA LUTESCENS* INFECTING THE HARLEQUIN LADYBIRD *HARMONIA AXYRIDIS* (COLEOPTERA: COCCINELLIDAE).

9:45 (76) † **René Monzalvo**, F Agustín Jiménez. PHYLOGENETIC AND MORPHOLOGICAL RELATIONSHIPS FROM *TRICHURIS* (NEMATODA: TRICHURIDAE) PARASITES OF RODENTS IN THE AMERICAS REVEAL INDEPENDENT LINEAGES.

7:50–10:00 am Special Symposium: Confluence A/B
Host-Parasite Interactions in Marine Ecosystems I

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding:	**Pieter Johnson,** University of Colorado, Boulder, CO **Dana Calhoun,** University of Colorado, Boulder, CO
7:50	Opening Remarks
8:00 (77)	**Kevin Lafferty**. MARINE DISEASES: AN OVERVIEW.
8:15 (78)	**Maureen A Williams**, Katie L Leslie, Emily Oven, Sara Faiad, Danielle Claar, Fiorenza Micheli, Ana S Guerra, Beverly French, Brian J Zgliczynski, Alison J Haupt, Stuart A Sandin, Chelsea L Wood. PARASITE LIFE HISTORY INTERACTS WITH LARGE SCALE DRIVERS OF PARASITE ABUNDANCE IN PACIFIC MARINE ECOSYSTEMS.
8:30 (79) †	**Hannah Bauman**, Lauren E Nadler, Jeffrey Hoch, Christopher Blanar. *EUHAPLORCHIS* SP. EFFECT ON SOCIAL BEHAVIOR AND FAMILIARITY OF GULF KILLIFISH (*FUNDULUS GRANDIS*).
8:45 (80)	**Pieter Johnson**, Remon Malawauw, Dana Calhoun, Zachary Kohl, Derek Zelmer. EMERGENCE OF *SCAPHANOCEPHALUS* INFECTIONS IN CARIBBEAN REEF FISH: EVIDENCE FROM MUSEUM SAMPLES AND CONTEMPORARY SAMPLING.
9:00 (81)	**Sean A Locke**, Dana M Calhoun, José M Valencia Cruz, Erika Taylor Ebbs, Vasyl V Tkach, Pieter Johnson. EXPANDING ON EXPANSUS: A NEW SPECIES OF *SCAPHANOCEPHALUS* FROM NORTH AMERICA AND THE CARIBBEAN BASED ON MOLECULAR AND MORPHOLOGICAL DATA.
9:15 (82) †	**Megan M Tomamichel**, Tina Walters, Elianna M K Fox, Ashly Rivera, Anita Minniefield, Max Braun, Richard J Hall, Marc E Frischer, James E Byers. CHANGES IN TRANSMISSION RATES DRIVE SEASONAL PATTERNS OF SHRIMP BLACK GILL DISEASE CAUSED BY THE PARASITIC APOSTOME CILIATE *HYALOPHYSA LYNNI.*
9:30 (83)	Rachel Goldberg, **Janet Koprivnikar.** DO CERCARIAE OF TREMATODE SPECIES FOUND IN DIFFERENT INTERTIDAL HABITATS SHOW CONSISTENCY IN KEY MORPHOLOGICAL TRAITS?
9:45 (84)	**John P McLaughlin**. PARASITES MAKE IMPORTANT AND CONSISTENT CONTRIBUTIONS TO TEMPERATE AND TROPICAL MARINE SYSTEMS.

10:00–10:20 am Coffee Break Mezzanine Foyer

10:20 am–Noon Student's Symposium Confluence C

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Haley R. Dutton,** Auburn University, Auburn, AL

10:20 Opening Remarks

10:30 (85) **Micah D. Bakenhaster**. PARASITES AND PUBLIC SERVICE: RESEARCH VIGNETTES FROM THE FLORIDA FISH AND WILDLIFE RESEARCH INSTITUTE.

11:00 (86) **Alistair DM Dove**. THE JOURNEY FROM PLANARIAN TO MULTIDISCIPLINARIAN.

11:30 Discussion and Concluding Remarks

10:20 am–Noon Special Symposium: Confluence A/B
Host-Parasite Interactions in Marine Ecosystems II

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Dana Calhoun,** University of Colorado, Boulder, CO
Pieter Johnson, University of Colorado, Boulder, CO

10:20 (87) **Christopher A Blanar**, Delaney Farrell, Hannah Bauman, Laura Nicolas, Nancy F Smith, Lauren E Nadler. EXPLORING PARASITE-INDUCED ALTERATIONS IN SOCIALITY AND BEHAVIOR IN ESTUARINE FISHES.

10:35 (88) **Dana M Calhoun**, Tyler J Achatz, Stephen E Greiman, Jasmine A Groves, John M Kinsella, Kara J Cromwell, Sean A Locke, Pieter T J Johnson. SPECIALIST WITHIN A SPECIALIST: INVESTIGATING INFECTION PATTERNS OF HELMINTH COMMUNITIES IN OSPREY ACROSS NORTH AMERICA WITH AN EMPHASIS ON *SCAPHANOCEPHALUS* SPP.

10:50 (89) **Eunji Park**, Brian Leander. PARASITES OF PARASITES: THE DIVERSITY OF POLYCHAETE-GREGARINE-METCHNIKOVELLID ASSOCIATIONS IN THE PACIFIC NORTHWEST.

11:05 (90) **Anindo Choudhury**, Alistair Dobson, Olivia Milloway, Gustavo A Castellanos-Galindo, Mark E Torchin. MARINE PARASITES AS COLONIZERS IN WAITING: INSIGHTS FROM THE PANAMA CANAL AS A NATURAL EXPERIMENT ON BIOLOGICAL INVASIONS.

11:20 (91) **Steven P Ksepka**, Alisa L Newton, Stephen A Bullard. DID BRAIN-INFECTING SCUTICOCILIATES (PHILISTERIDA SP.) KILL WILD SHARKS IN THE NORTHWESTERN ATLANTIC OCEAN?

11:35 (92) † **Xuqing Chen**, Kimberly S Reece, Jeffrey D Shields. DIFFERENT LIFE HISTORY STRATEGIES IN THE TRANSMISSION OF *HEMATODINIUM PEREZI* (DINOFLAGELLATA: SYNDINIALES) FROM THE BLUE CRAB (*CALLINECTES SAPIDUS*).

11:50 Discussion and Concluding Remarks

Noon–1:30 pm Students' Luncheon & Vortex Platte River

All students are invited to lunch. Non-students are by invitation only. Students will have an opportunity to socialize, network, and ask questions with a mix of principal investigators.

1:30–3:30 pm Host-Parasite Interactions II Confluence C

Time (Abstract No.)

† denotes student presentation in the Best Student Presentation Competition

Presiding: **Jasmine Groves**, University of Colorado, Boulder, CO

1:30 (93) Rachel Horjesi, Chase Nelson, Avery De Ruyter, Helen Gensch, Carly Weber, Maggie Bagatta, **Nicolas J Wheeler.** QUANTITATIVE ETHOLOGY OF SCHISTOSOME MIRACIDIA AND OTHER AQUATIC INVERTEBRATES.

1:45 (94) † **Sidney C Mann**, Jillian T Detwiler. NO DETECTION BIAS BETWEEN TWO SAMPLING APPROACHES FOR INTERMEDIATE HOSTS INFECTED WITH BRAINWORM (*PARELAPHOSTRONGYLUS TENUIS*).

2:00 (95) † **Brendan K Hobart**, Wynne E Moss, Travis McDevitt-Galles, Tara E Stewart Merrill, Pieter TJ Johnson. IT'S A WORM-EAT-WORM WORLD: CONSUMPTION OF TREMATODE (DIGENEA) FREE-LIVING STAGES PROTECTS SNAIL HOSTS AND BENEFITS ANNELID PREDATORS.

2:15 (96) † **Kevin A McQuirk**, Juliana M DeCore, Tathagata Debnath, Donald O Natvig, Maria G Castillo, Coen M Adema. DIGENEAN TREMATODES ASSOCIATED WITH HYGROPHYLID SNAILS AT THE RIO GRANDE NATURE CENTER (NM) WITH SPECIAL CONSIDERATION OF *PHYSELLA ACUTA* SNAILS.

2:30 (97) Jenna D Kirkland, Katherine Orndorff, Roger Ramirez Barrios, Anne C Jones, Mason C Martin, Tappey H Jones, **Ashleigh B Smythe**. TENEBRIONID BEETLES SHOWS ATTRACTION TO SOME, BUT NOT ALL, ACIDS FOUND IN THE FECES OF RATS INFECTED WITH THE TAPEWORM *HYMENOLEPIS DIMINUTA.*

2:45 (98) Ryan W Koch, **Matthew G Bolek**. LIFE CYCLE VARIATION AND HOST-PARASITE INTERACTIONS OF FOUR CONGENERIC SPECIES OF TURTLE ACANTHOCEPHALANS.

3:00 (99) Lijun Lu, Lijing Bu, Martina R Laidemitt, Zhang Si-Ming, **Eric S Loker**. DIFFERENT METAZOAN PARASITES, DIFFERENT TRANSCRIPTOMIC RESPONSES, WITH NEW INSIGHTS ON PARASITIC CASTRATION BY DIGENETIC TREMATODES IN THE SCHISTOSOME VECTOR SNAIL *BIOMPHALARIA GLABRATA.*

3:15 (100) **Tom Pennance**, Jacob A Tennessen, Johannie M Spaan, Tammie McQuistan, George Ogara, Fredrick Rawago, Martin Mutuku, Gerald M Mkoji, Eric S Loker, Maurice Odiere, Michelle L Steinauer. GENOME WIDE ASSOCIATION STUDY OF AN AFRICAN SNAIL VECTOR OF SCHISTOSOMIASIS IDENTIFIES GENES ASSOCIATED WITH RESISTANCE TO INFECTION BY *SCHISTOSOMA MANSONI.*

1:30–3:30 pm Taxonomy, Systematics, & Phylogeny II Confluence A/B

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Spencer Galen,** University of Scranton, Scranton, PA

1:30 (101) † **Zoe S Von Holten**, Tyler J Achatz, Vasyl V Tkach. SSS-SLIPPERY SYSTEMATICS OF SLITHERING SNAKE WORMS.

1:45 (102) † **Kamila Cajiao-Mora**, John H Brule, Haley R Dutton, Stephen A Bullard. SUPPLEMENTAL DESCRIPTION OF *CABALLEROTREMA ANNULATUM* (DIESING, 1850) OSTROWSKI DE NÚÑEZ AND SATTMANN, 2002 (DIGENEA: CABALLEROTREMATIDAE) FROM A NEW HOST (*ELECTROPHORUS* CF. *VARII*) AND LOCALITY (AMAZON RIVER, COLOMBIA) WITH PHYLOGENETIC ANALYSIS AND EMENDED GENERIC DIAGNOSIS.

2:00 (103) † **Maggie A Young**, Sarah A Orlofske, Robert C Jadin, Stephen E Greiman, Vasyl V Tkach, Tyler J. Achatz. GUT SECRETS OF RACCOONS: UNVEILING ENIGMATIC NEW SPECIES OF ALARIA.

2:15 (104) † **Morgan B Fleming**, Florian B Reyda. A DEEPER LOOK AT HOOKS: INTER-RELATIONSHIPS AMONG NEOECHINORHYNCHID ACANTHOCEPHALANS.

2:30 (105) † **John H Brule**, Micah B Warren, Stephen A Bullard. TWO AND A HALF CENTURIES OF MORPHOLOGICAL MISHAPS WITHIN *AXINE* ABILDGAARD, 1794 (MONOGENOIDEA: AXINIDAE MONTICELLI, 1903): NEW OBSERVATIONS OF TYPE SPECIMENS, DESCRIPTION OF A PUTATIVE INNOMINATE SPECIES, AND (FINALLY) A TREE THAT INCLUDES AN AXINID SEQUENCE.

2:45 (106) † **Jessica L Rotolo**, John R Barta. COCCIDIAL DESCRIPTIONS IN THE MOLECULAR AGE – WITH A FOCUS ON *EIMERIA* SPECIES.

3:00 (107) † **Allison Bryant**, Matthew Bolek. PURCHASING YOUR FIRST PET COCCIDIAN: THE DIVERSITY OF COCCIDIA AND RELATED PARASITES (APICOMPLEXA) FROM PET STORE SQUAMATE REPTILES.

3:15 (108) † **Kapepula Kasembele Gyrhaiss**, Maarten Vanhove. DIVERSITY OF *QUADRIACANTHUS* PAPERNA, 1961 (MONOGENEA: DACTYLOGYRIDAE) IN THE UPPER CONGO BASIN: NEW GEOGRAPHICAL RECORDS AND DESCRIPTION OF FIVE NEW SPECIES FROM THE GILLS OF *CLARAIS NGAMENSIS* CASTELNAU, 1861 (SILURIFORMES: CLARIIDAE).

1:30–3:30 pm Ecology & Evolution II Lawrence A/B

Time (Abstract No.)
† denotes student presentation in the Best Student Presentation Competition

Presiding: **Michael R Zimmermann,** University of Mount Union, Alliance, OH

1:30 (109) † **Taylor M Fiedor**, Maria R Bollinger, Samuel N Messinides, Kyle D Gustafson. MONTANE MOLLUSCS: FACTORS AFFECTING SNAILS AND THEIR TREMATODES THROUGHOUT THE MOUNTAIN ECOREGIONS OF ARKANSAS.

1:45 (110) † **Jenna M Hulke**, Charles D Criscione. TESTING EVOLUTIONARY THEORY INTERSECTING COMPLEX LIFE CYCLES AND HERMAPHRODITIC MATING SYSTEMS FROM FIELD-COLLECTED PARASITES.

2:00 (111) † **Lauren Edison**, Cameron P Goater. CHANGES IN ENVIRONMENTAL CONDITIONS DETERMINE A BIZARRE HOST MANIPULATION IN CANADA'S FLUKE-INFECTED ZOMBIE ANTS.

2:15 (112) **Kyle D Gustafson**, Taylor M Fiedor. FOLLOWING THE SNAIL TRAIL: UNCOILING THE FACTORS THAT DRIVE THE DISTRIBUTIONS OF TREMATODES AMONG MULTIPLE ECOREGIONS.

2:30 (113) † **Monica Reusche**, Hannah Warr, Kyle D Gustafson, Ronald Johnson, Virginie Rolland, Andrew D Sweet. SCRATCHING AT THE PHYLOGENY OF THE POCKET GOPHER LICE: ASSESSMENT FOR *GEOMYS BURSARIUS OZARKENSIS.*

2:45 (114) † **Dani R Jakovljevic**, Alyssa Turnbull, Chloe Christenson, Patrick Hanington. FINDINGS ON *MYXOBOLUS CEREBRALIS* IN NEWLY INVADED ALBERTA, CANADA.

3:00 (115) † **Joshita Sehgal**, Harold M Aukema, Jillian T Detwiler. DETERMINING THE OPTIMAL DENSITY OF CERCARIAE FOR PARASITE OXYLIPIN PROFILES.

3:15 (116) **Antoine Filion**, Sundaram Mekala, John Paul Schmidt, John M Drake, Patrick R Stephens. THOSE WHO FORGET THE PAST ARE CONDEMNED TO REPEAT IT: EVIDENCE OF REPEATED ZOONOTIC PATHOGEN SPILLOVER AT ECOLOGICAL BOUNDARIES.

3:30–4:30 pm Students' Business Meeting Platte River

Presiding: **Haley R Dutton**, Auburn University, Auburn, AL

4:30–6:30 pm Poster Session and Palettes & Parasites Molly Brown & Horace Tabor

(Snacks and Beverages)

† denotes student presentation in the Best Student Presentation Competition

CELL BIOLOGY

117 † **Jasper C Rowe.** TRACKING EXTRACELLULAR VESICLE RELEASE IN NEMATODES.

CHEMOTHERAPY & DRUG RESISTANCE

118 Olubukola Owolodun, Benson Otarigho, **Mofolusho Falade**. *COSTUS AFER* AND *COSTUS SPECTABILIS* ENHANCE SURVIVAL AND PROMOTE HOST IMMUNE RESPONSE AGAINST *PLASMODIUM BERGHEI* INFECTION IN MICE.

119 **Sophie Willett**, Zachary Caterer, Sage Mathisen, Nicolas J Wheeler**.** WRMXPRESS GUI: A USER-FRIENDLY INTERFACE FOR HIGH-THROUGHPUT ANALYSIS OF PARASITIC WORMS.

ECOLOGY & EVOLUTION

120 † Taylor M Fiedor, **Samuel N Messinides**, Faith Stephens, Kyle D Gustafson. SEQUENCING SNAILS AND THEIR TREMATODE CERCARIAE REVEALS HOST SPECIFICITY PATTERNS AND CRYPTIC DIVERSITY AMONG MULTIPLE ECOREGIONS.

121 † **Laura A Redfield**, Graham B Goodman. COMPARATIVE PRESERVATION METHODS FOR PARASITES IN FECAL SAMPLES: VACUUM SEALING VS. CONVENTIONAL STORAGE.

122 † **Sophia R Holupka**, Ramon A Carreno. PARASITE COMMUNITIES IN SUNFISH (CENTRARCHIDAE) FROM THE OLENTANGY RIVER, OHIO, USA.

123 † **Anthony Raona**, Michael Joyce, Joseph Jacquot, Paul Keenlance. PARASITE MEDIATED COMPETITION BETWEEN FLYING SQUIRRELS (*GLAUCOMYS*).

124 † **Josh Pletcher**, Cassie Stitzman, Kurt Galbreath. FLEA FAUNA OF MONGOLIAN PIKAS, JERBOAS, AND ZOKORS.

125 **Emily Greenman**, Josh Sharp, Kurt Galbreath. *BARTONELLA* DIVERSITY IN SMALL MAMMALS OF MONTANA AND IDAHO.

126 **Sarah A Orlofske**. DIGITIZING A PARASITE COLLECTION AT A PRIMARILY UNDERGRADUATE INSTITUTION: CHALLENGES AND OPPORTUNITIES.

127 **Anindo Choudhury**, Michelle Steinauer, Jayde A Ferguson, Michael L Kent. PARASITES OF FISHES FROM THE GLADSTONE AND ISAAC-SEKULMUN LAKES IN THE YUKON: IMPLICATIONS FOR A PROPOSED WATER DIVERSION AND HYDROELECTRIC POWER PROJECT.

128 **Nicholas C Tsangarides**, Jade Salis, Whitney C Preisser. BENEATH THE SURFACE: UNDERSTANDING PARASITIC FAUNA IN DAM-ALTERED ECOSYSTEMS.

129 † **Katherine Garcia**, Sarah Orlofske. ECTOPARASITE COMMUNITIES IN BARRED OWLS (*STRIX VARIA*) USING NEW AND ARCHIVED MUSEUM COLLECTIONS.

130 † **Roxanne M Gasperetti**, Rianna J Taylor, Vasyl V Tkach, Tyler J Achatz, Jeffrey A Bell, Mike Kinsella, Sarah A Orlofske. PARASITES OF THE AMERICAN WOODCOCK (*SCOLOPAX MINOR*) FROM WISCONSIN, USA.

131 † **Connor McCowan**, Christina Anaya. PROPOSED PROJECT: CAN A NATIVE TOAD, *ANAXYRUS TERRESTRIS, BECOME INFECTED* WITH THE NONNATIVE NEMATODE, *RHABDIAS PSEUDOSPHAEROCEPHALA.*

132 **Andreas Schmidt-Rhaesa**, Andreas Hejnol, Maria Weißbecker, Shumpei Yamakawa. A NOVEL FORM OF EGGSTRINGS FOR EUROPEAN NEMATOMORPHS.

133 **Andreas Schmidt-Rhaesa**, Andreas Hejnol. COLLECTING *NECTONEMA*, THE MARINE NEMATOMORPHA.

134 **Andreas Schmidt-Rhaesa**, Andreas Hejnol. REMARKS ON INDIAN GORDIIDS (NEMATOMORPHA).

GENOMICS & MOLECULAR BIOLOGY

135 Md Makshuder Rahman Zim, **Md Mahfuzur Rahman Sajib**, Nurnabi Ahmed, Md Rajiur Rahman Rabbi, Mostak Ahmed, Md Khalilur Rahman, Babul Chandra Roy, Md Hasanuzzaman Talukder. SEROPREVALENCE AND MOLECULAR CHARACTERIZATION OF BOVINE ANAPLASMOSIS IN BANGLADESH.

136 **Matthew S Tucker**, Celia N O'Brien, Alexis N Johnson, Benjamin M Rosenthal, Mark C Jenkins. TRANSCRIPTIONAL PROFILES OF SPORULATION IN SPECIES OF *EIMERIA* INFECTING CHICKENS.

HOST-PARASITE INTERACTIONS

137 **Abdulaziz Alouffi**. *FASCIOLA*-INFILTRINS AS A TARGET FOR VACCINATION.

138 **David J Blake**, Jaslin Lindsay, Kiara Nanez, Kenny Miller. UTILIZING NATURAL PRODUCTS TO INHIBIT LEISHMANIA GROWTH AND AMASTIGOTE FORMATION BY MODIFYING ESSENTIAL HOST PATHWAYS IN HUMAN MACROPHAGES.

139 † **Sofiane Aiche**. MOLECULAR INVESTIGATION OF TICK-BORNE PATHOGENS IN FERAL SWINE (*SUS SCROFA*) FROM SEVEN STATES IN USA.

140 † Sydney Watson, **Jillian Vallance**, Dana M Calhoun, Pieter TJ Johnson. QUANTIFYING PARASITE-INDUCED SEVERITY OF BLACK SPOT SYNDROME IN *ACANTHURUS TRACTUS* ON REEF SITES IN BONAIRE.

141 **Alyssa O'Malley**, Alexandra R Williams, Nina Rehfeldt, Shreeya Mishra, Scott Ngyuen, Jack Lundstrom, John Shea. *MELITTOBIA DIGITATA* PARASITOIDS AND HOST PREFERENCE – EXAMINING HOW ABSENCE OF PREFERRED HOSTS AFFECTS HOST SELECTION.

142 † **Rachel Horejsi**, Chase Nelson, Ireland Coughlin, Carly Weber, Nicolas J Wheeler. DEVELOPING METHODS TO QUANTIFY *SCHISTOSOMA MANSONI* MIRACIDIA BEHAVIOR AND RESPONSIVENESS.

143 **Hannah Bialic**, Laura Knoll, David Beebe, Marcos Lares, Kehinde Adebayo Babatunde. INTERROGATION OF *ENTAMOEBA HISTOLYTICA* IN A HUMAN INTESTINAL TISSUE MICROPHYSIOLOGICAL SYSTEM.

144 † **Jade Salis**, Whitney Preisser. ASSESSING RELATIONSHIPS BETWEEN MICROPLASTIC ABUNDANCE AND PARASITE LOAD IN FRESHWATER FISH FROM RACCOON CREEK AND PUMPKINVINE CREEK, GEORGIA, USA.

145 † **Md Rowshan Alam,** Elliot Zieman, Robert Colombo. PARASITE PREVALENCE DIFFERS IN BLUNTNOSE MINNOWS (*PIMEPHALES NOTATUS*) ACROSS CENTRAL ILLINOIS CREEKS.

146 † **Madison Ustohal**, Alyssa Bolger, John Shea**.** BEHAVIORAL CHANGES IN CRICKETS INFECTED WITH *PARAGORDIUS VARIUS.*

IMMUNOLOGY

147 Christopher J Brianik, Younes Bouallagui, **Bassem Allam**. THE EFFECT OF PLOIDY ON THE FORMATION AND ANTIMICROBIAL EFFICACY OF EXTRACELLULAR TRAPS IN THE EASTERN OYSTER.

148 † **Kacey Newhart**, Elliott Zieman. FELINE CYTOKINE RESPONSE TO *CYTAUXZOON FELIS* INFECTIONS WITH DIFFERENT CLINICAL PRESENTATIONS.

LIFE CYCLES & EPIDEMIOLOGY

149 **Chad L Cross**, Bryson Carrier, Miklo Alcala, Louisa A Messenger. SOIL-TRANSMITTED HELMINTHS IN THE UNITED STATES: USING BIG DATA TO CHARACTERIZE PATIENTS AND ANALYZE DISEASE TRENDS.

150 † **Daniel F Griesemer**, John Shea. TESTING THE PRESENCE OF PHEROMONES IN HORSEHAIR WORMS (*PARAGORDIUS VARIUS*) USING AN ARTIFICIAL STREAM.

151 **Thomas B Hayssen**, Sarah Herzer, Noah Takamiya, John Shea. INVESTIGATIVE STUDY INTO REARING *CHORDODES MORGANI* IN A LAB.

152 **Alexander D Hernandez**, Sara Major. RELATING HOG MANAGEMENT PRACTICES TO NEMATODE INFECTION ON ORGANIC AND PASTURED PIG FARMS.

153 **Christina Yeager**, Matthew Tucker, Alan Gutierrez, Celia O'Brien, Manan Sharma, Valsin Fournet, J P Dubey, Mark Jenkins, Kali Kniel, Benjamin M Rosenthal. A MINIATURIZED SAND-ZERO VALENT IRON (ZVI) FILTER EFFECTIVELY TRAPS THE COCCIDIAN PARASITES *C. CAYETANENSIS* AND *EIMERIA*: IMPLICATIONS FOR RISK MITIGATION IN IRRIGATION WATER.

154 † **Ashley G McDonald**, Jennifer R Schultze, Clayton K Nielsen, F Agustin Jimenez. PREVALENCE AND DISTRIBUTION OF ZOONOTIC PATHOGENS AND PARASITES IN RACCOONS (*PROCYON LOTOR*) OF NORTHERN ILLINOIS.

TAXONOMY, SYSTEMATICS, & PHYLOGENY

155 † **Katherine M Hanselman**, Kirsten Jensen. THE PHYLOGENETIC PLACEMENT OF LECANICEPHALIDEANS PARASITIZING THE COWTAIL STINGRAY GENUS *PASTINACHUS.*

156 † **Katerina D Sawickij**, Madison Stanley, Florian Reyda. FINDING DORY (*NEOECHINORHYNCHUS DORYPHORUS*): VAN CLEAVE AND BANGHAM’S MYSTERY WORM REDISCOVERED IN THE FLORIDA EVERGLADES.

157 **Allison Bryant**, Matthew Bolek, Gabriel Langford. NEW INSIGHTS INTO REPTILIAN COCCIDAN INFECTIONS FROM TWO SPECIES OF INVASIVE GECKOS, THE MEDITERRANEAN HOUSE GECKO, *HEMIDACTYLUS TURCICUS* AND THE TROPICAL HOUSE GECKO, *H. MABOUIA* FROM THE NEW WORLD

158 Ryan P Shannon, Chris T McAllister, **Matthew G Bolek**. MORPHOLOGICAL AND MOLECULAR CHARACTERIZATION OF *HEPATOZOON* (APICOMPLEXA: ADELEORINA) SPECIES INFECTING FROGS AND SNAKES ACROSS THE CENTRAL AND EASTERN UNITED STATES.

159 **Timothy F Picozzi**, Jessica E Ozner, Florian B Reyda. ON THE IDENTITY OF A PROBLEMATIC SET OF *NEOECHINORHYNCHUS* SPECIMENS FROM BUFFALO (CATOSTOMIDAE) FROM ILLINOIS.

VECTOR BIOLOGY

160 **Avery De Ruyter**, Helen Gensch, Maggie Bagatta, Nicolas J Wheeler. CHEMOSENSORY BEHAVIORS OF LARVAL *AEDES TRISERIATUS* MOSQUITOS.

161 **Cassie M Stitzman**, Kurt E Galbreath. INVESTIGATING *BARTONELLA* PREVALENCE OF INTERMOUNTAIN WEST FLEAS.

Monday, June 17

8:15–10:00 am Taxonomy, Systematics, & Phylogeny III Platte River

Time (Abstract No.)
Presiding: **Stephen A Bullard**, Auburn University, Auburn, AL

8:15 (162) **Micah B Warren**, Steven P Ksepka, Triet N Truong, Stephen S Curran, Haley R Dutton, Stephen S Bullard. ANOTHER NEW GENUS AND SPECIES OF FISH BLOOD FLUKE (DIGENEA: APOROCOTYLIDAE) FROM A COMMON FISH (WHITE MULLET, *MUGIL CUREMA*) IN GULF OF MEXICO, INCLUDING PATHOLOGY AND PHYLOGENETIC ANALYSIS.

8:30 (163) **Stephen S Curran**, Steven P Ksepka, Stephen A Bullard. PHYLOGENETICS OF TWO “RELATED” MONOTYPIC DIGENEANS, *MICROCREADIUM PARVUM* SIMER, 1929 AND *POSTPORUS EPINEPHALI* (MANTER, 1947) MANTER, 1949.

8:45 (164) **Triet N Truong**, Stephen A Bullard. *DULCITRANSVERSOTREMA* N. GEN. (DIGENEA: TRANSVERSOTREMATIDAE) AND FIRST MORPHOLOGICAL AND NUCLEOTIDE-BASED CONFIRMATION OF THE LIFE CYCLE OF A TRANSVERSOTREMATID IN NORTH AMERICA.

9:00 (165) **Tyler J Achatz**, Francisco TV Melo, Martin M Montes, Vasyl V Tkach. WHAT CONTINENTAL DRIFT? PHYLOGENY, BIOGEOGRAPHY AND HOST ASSOCIATIONS OF THE PROTERODIPLOSTOMIDAE.

9:15 (166) **Erin L Hobday**, Dana M Calhoun, Tyler J Achatz, Pieter TJ Johnson. UNDER THE SURFACE: UNDERSTANDING INFECTION PATTERNS AND MORPHOLOGY OF *HAEMATOLOECHUS* SPP. IN CALIFORNIA BULLFROGS.

9:30 (167) **Vasyl V Tkach**, Tyler J Achatz, Aneta Kostadinova, Simona Georgieva, Alan Fecchio. MOLECULAR PHYLOGENY REVEALS HISTORY OF HOST SWITCHING EVENTS IN THE EVOLUTION OF *NEODIPLOSTOMUM* (DIGENEA, DIPLOSTOMIDAE).

9:45 (168) **F Agustín Jiménez**. A PLEA TO DEVELOP A CONVENTION TO SHARE DATASETS THROUGH UNIVERSAL DATA REPOSITORIES.

8:00–10:00 am Host-Parasite Interactions III Molly Brown & Horace Tabor

Time (Abstract No.)

Presiding: **Patrick Hanington,** University of Alberta, Edmonton, Canada

8:00 (169) Dana M Calhoun, **Jasmine A Groves**, Paula A Schaffer, Tyler J Achatz, Stephen E Greiman, John M Kinsella, Pieter TJ Johnson. TIGERS WITH SPOTS: SEVERE CLINOSTOMID OUTBREAK WITH HIGH MORBIDITY IN *AMBYSTOMA TIGRINUM* FROM BOULDER, COLORADO.

8:15 (170) **Md Rowshan Alam**, Elliot Zieman, Robert Colombo. PARASITE PREVALENCE DIFFERS IN BLUNTNOSE MINNOWS (*PIMEPHALES NOTATUS*) ACROSS CENTRAL ILLINOIS CREEKS.

8:30 (171) **Graham B Goodman**, Hannah Huston, Devon Adamo, Ruben F Navarro, Bradley J Cosentino. EFFECT OF COAT COLOR ON PARASITISM IN EASTERN GRAY SQUIRRELS.

8:45 (172) **Laila Brubaker**, Ben Americus, Golan Nadav, Tamar Lotan, Jerri Bartholomew, Stephen Atkinson. DISRUPTING HOST SENSING OF THE MYXOZOAN PARASITE *CERATONOVA SHASTA.*

9:00 (173) **Margaret L Doolin**, M Denise Dearing. COULD PINWORMS BE MUTUALISTS IN FIBER DIGESTION?

9:15 (174) **Bassem Allam**, Denis Grouzdev, Stephen Tettelbach, Harrison Tobi, Arni Kristmundsson, Emmanuelle Pales Espinosa. CHARACTERIZATION OF AN APICOMPLEXAN PARASITE ASSOCIATED WITH THE COLLAPSE OF THE BAY SCALLOP (*ARGOPECTEN IRRADIANS*) POPULATION IN NEW YORK.

9:30 (175) **Marin Milotic**, Alan J Lymbery, Andrew R C Thompson, Leighton J Thomas, Adria R A Eda, Stephanie S Godfrey. PARASITE COMMUNITIES OF CRITICALLY ENDANGERED HOSTS ARE INFLUENCED BY DIFFERENT SITES OF TRANSLOCATION AS REVEALED BY METABARCODING.

9:45 (176) **Mohammad K Saifullah**, Anam Sahreen. ANTHELMINTIC EFFICACY OF *TRIGONELLA FOENUM GRAECUM* SEED EXTRACTS AGAINST *ISOPARORCHIS HYPSELOBAGRI* (DIGENEA: TREMATODA) INFECTING SWIM BLADDER OF FRESHWATER CAT FISH *WALLAGO ATTU*.

8:00–10:00 am Ecology & Evolution III Lawrence A/B

Time (Abstract No.)

Presiding: **Sarah A Orlofske,** University of Wisconsin-Stevens Point, Stevens Point, WI

8:00 (177) Colin C MacLeod, **Lien T Luong**. ECOLOGY OF FEAR: *DROSOPHILA NIGROSPIRACULA* PRIORITIZES ANTI-PREDATOR BEHAVIOUR AT THE EXPENSE OF SUB-LETHAL PARASITIC INFECTION.

8:15 (178) **Janine N Caira**, Kirsten Jensen. TEENY, TINY, TAPEWORMS—THEY FIT ON THE HEAD OF A PIN.

8:30 (179) **Armando J Cruz-Laufer**, Maarten P M Vanhove, Lutz Bachmann, Christoph Hahn, Philipp Resl, Nikol Kmentová. ADAPTIVE EVOLUTION OF STRESS RESPONSE GENES IN TWO MONOGENEAN PARASITE LINEAGES ALIGNS WITH THEIR RESPECTIVE HOST NICHE DIVERSITY.

8:45 (180) **Michael R Zimmermann**, Cassidy Wells, Simone Meadows. VARIATION IN DIPLOSTOMID PARASITE INFECTION PATTERNS AND PATHOLOGY IN BLUEGILL SUNFISH (*LEPOMIS MACROCHIRUS*) REPRODUCTIVE MORPHOTYPES.

9:00 (181) **Michael R Zimmermann**, Madison Upperman. THE ROLE OF CENTRACHID HOST DIVERSITY ON *POSTHODIPLOSTOMUM* SPP. INFECTION IN BLUEGILL SUNFISH (*LEPOMIS MACROCHIRUS*).

9:15 (182) Sarah E Unrau, **Cameron P Goater**. VISUALIZATION OF THE HOST-PARASITE INTERFACE UNCOVERS POTENTIAL MECHANISMS OF BEHAVIOR MANIPULATION IN *DICROCOELIUM*-INFECTED ZOMBIE ANTS.

9:30 (183) **Matthew G Bolek**, Ryan P Shannon, David D Berman, Kristen A Baum. EVOLUTION OF A PATHOGENIC NEOGREGARINE (*OPHRYOCYSTIS ELEKTROSCIRRHA*) IN MILKWEED BUTTERFLIES.

9:45 (184) Pablo Oyarzún-Ruiz, Richard Thomas, Adriana Santodomingo, Martín Zamorano-Uribe, Manuel Moroni, Lucila Moreno, Sebastián Muñoz-Leal, Verónica Flores, **Sara V Brant**. AVIAN SCHISTOSOMATIDS AND THEIR LIFE CYCLES IN SOUTHERN CONE OF SOUTH AMERICA.

10:00–10:20 am Coffee Break Mezzanine

10:20–11:20 am Eminent Parasitologist Lectureship Award Confluence A/B/C

Presiding: **Susan Perkins,** The City College of New York, New York, NY

10:20 am **Susan Perkins**. Introduction of the Eminent Parasitologist Lectureship Award.

10:30 am (185) **Robert Poulin**. “An Ecologist’s Journey in Parasite Land.”

1:00–2:00 pm H. B. Ward Medal Lecture Confluence A/B/C

Presiding: **Kristin Herrmann,** Tarleton State University, Stephenville, TX

1:00 pm **Pieter Johnson**. Introduction of the 2024 H. B. Ward Medal Recipient.

1:10 pm **Janet Koprivnikar**. "A MacGyver Approach to Parasitology."

Dr. Janet Koprivnikar. Recipient of the 2024 H. B. Ward Medal.

2:00–4:30 pm ASP Awards & Business Meeting Confluence A/B/C

ASP Awards:

Presiding: **Kristin Herrmann,** Tarleton State University, Stephenville, TX

Clark P. Read Mentor Award Lecture

2:00 pm **Jeffrey Bell**. Introduction of the 2024 Clark P. Read Mentor Award recipient.

2:10 pm **Vasyl Tkach.**

Dr. Vasyl Tkach. Recipient of the Clark P. Read Mentor Award.

Monday, June 17th

Ashton Cuckler New Investigator Award

Presiding: **Kristin Herrmann,** Tarleton State University, Stephenville, TX

Dr. Kaylee Herzog. Recipient of the Ashton Cuckler New Investigator Award.

Best Student presentations and Marc Dresden Travel Grant Awards

Presiding: **Christina Anaya**, Florida Gulf Coast University, Fort Myers, FL

ASP Business Meeting

Presiding: **Reginald Blaylock,** University of Southern Mississippi**,** Ocean Springs, MS

Thank you for participating and making this another successful ASP meeting.

See you all next year 7-10th July in Winston-Salem, NC

Abstracts

1

Integrating active learning strategies into parasitology courses to promote student-centered learning

Christina Anaya
Florida Gulf Coast University, Fort Myers, FL, USA.

An active learning pedagogy attempts to engage students by discussing, thinking, writing, talking, interpreting, investigating, and creating during class time. Therefore, using an active-learning pedagogy employs a variety of methodologies. During this time, the instructor fits a variety of profiles including as a subject expert during traditional lectures but as a facilitator while engaging students in active learning strategies. These profiles can also include coach and evaluator and an engaging classroom often sees transitions between the profiles. By designing learning activities around learning outcomes, students become accountable to participate in and outside of the classroom and become more engaged when less traditional lectures are presented. In this presentation, I will demonstrate active learning strategies that employs development of course content and learning activities all in a parasitological context. Attendees will be given the opportunity to demonstrate and create activities during the active learning session.

2

Flipping out: Opportunities and Challenges for Parasitology in a “Flipped” Classroom

Sarah A Orlofske
University of Wisconsin - Stevens Point, Stevens Point, WI, USA.

Flipped classrooms, a teaching environment where students watch interactive online videos and/or recorded lectures, open classroom time for discussion and exploring specific concepts in greater depth. Flipped classrooms have many benefits for instructors including recognizing student struggles with “homework”, using classroom time more effectively and creatively, and providing flexibility for students who miss class due to illness, sports, or other extracurricular activities. The benefits to student learning outcomes, active engagement, and enjoyment have made the method attractive to instructors in a variety of science disciplines. Using a flipped classroom for parasitology can provide benefits such as additional time with scientific equipment like microscopes only available in the classroom as well as time for students to spend on case studies and authentic research. In this presentation, I will explore potential opportunities for using the flipped classroom approach in an undergraduate parasitology course as well as explain the challenges of implementation and how to mitigate them. Specifically, I will highlight a series of active learning activities that are useful for in-class time freed by the flipped classroom approach. Examples of traditional activities include discussions of peer-reviewed scientific literature, case studies, and debates. However novel activities include a public outreach project where students choose a target audience and a taxonomic group of parasites or concept in parasitology and a creative mechanism to educate their public audience. Using in-class time for feedback, design, and background research combined with independent creativity of the students leads to unique games, displays, songs, and even baked goods that the students enthusiastically share with each other and the public through an annual behind the scenes museum tour. Additional activities include a Parasite Madness match up modeled after March Mammal Madness in a basketball bracket style tournament that requires students to research their combatants and use their teamwork and problem-solving skills to decide who will win. Originally developed by an undergraduate teaching assistant this fun and challenging activity demonstrates the freedom and originality afforded by the flipped classroom approach. By sharing flipped classroom teaching ideas, we can improve learning outcomes for students and inspire future parasitologists.

3

Incorporating technology supported, team-based learning into a Parasitology course

Ana E Garcia Vedrenne
University of California, Irvine, Irvine, CA, USA.

Over the past years, there has been increased pressure in higher education to shift away from traditional lectures as the dominant delivery mode. Here, I share components of an upper-division Parasitology course designed with the explicit purpose of building community and collaboration in the classroom through self-guided, team-based active learning. Students are assigned to their teams at the start of the quarter and equipped with various resources to help establish healthy team dynamics. Accountability for individual and group work is achieved through pre-class preparation, in-class structured group activities with defined roles, and individual reflections on team contributions. During class time, students collaboratively engage with prompts provided in Google slide templates. These templates aim to support students as they grasp key disciplinary concepts and foster an inclusive learning environment. Advantages of this format include that a) assignments promote collaborative learning, b) students develop important professional skills such as communication, teamwork, and time management, c) the structured learning provides a framework for a diverse body of instructors to effectively facilitate the activity (e.g., graduate teaching assistants with varying levels of teaching experience or expertise on the course topic), and d) it is scalable to large enrollment courses. I am excited to share the activities I developed and have ongoing discussions about best practices for creating environments conducive to learning and team development.

4

Assessment of Student Learning Outcomes and Perceptions of Active Learning in an Undergraduate Parasitology Course

Joseph T Cornelius[1], Joanna J Cielocha[2]
[1]Indiana University School of Medicine - Bloomington, Bloomington, IN, USA. [2]Rockhurst University, Kansas City, MO, USA.

Learning objectives are utilized in courses as an organizational means to content presentation and progress throughout the semester. We crafted meaningful learning objectives through the surveying and conflation of 20 undergraduate or graduate-level parasitology course syllabi available on the web. This resulted in the identification of 9 common parasitological themes: parasite distribution; ecology; morphology; evolution; life cycles; pathogenesis and disease; classification and taxonomy; environmental biology; economics; transmission; treatment; global and cultural impact; and host biology. Based on these objectives, a pre- and post-assessment was designed to measure student content mastery (i.e., end of semester) compared to prior knowledge (i.e., beginning of semester). The assessment was administered in an undergraduate parasitology course at a small liberal arts university in the midwestern United States in Fall 2021 (N=22) and Fall 2023 (N=18). Additionally, throughout the semester, students engaged in three active learning sessions. These sessions provided reflective based data points, giving insight into student comprehension beyond a summative measurement. Student surveys were administered at the mid-point of the semester and the end of the semester to gather feedback on the use of active learning tools in the course and to gain insight into student learning. In this multiple methods study both formative and summative assessment data was collected to understand student comprehension and learning related to the learning objectives. A thematic analysis was utilized on student reflection data to observe the relationship between student comprehension and active learning tools. Statistical methods were employed to analyze pre/post-test assessments and overall knowledge gaps in specific learning objectives. This study provided meaningful data in relation to learning objectives in an active classroom. Quantitatively, it provided evidence of significant gains with class averages on the pre-assessment of 58% and 48% on the pre-assessment vs. 91% and 86% on the post-assessment in 2021 and 2023, respectively. Through the qualitative analysis, multiple themes emerged in association with learning processes, active participation in class, study habits, and general trends in confidence when working with parasitology content. Overall, connecting student learning objectives with formative assessment allowed for measurable outcomes of student progress in course material.

5

Using the Imagination to Teach Parasitology with Case Studies and Creativity

John F Shea
Creighton University, Omaha, NE, USA.

Keeping the attention of undergraduates is challenging, especially when an increasing number of students suffer from sleep deprivation as well as mental health challenges such as anxiety, depression and ADD. Further, social media and online shopping compete for the instructor's attention. Engaging student imagination can help overcome these challenges. I describe two ways to engage imagination through case studies and creativity. Many students taking Parasitology are in a pre-health track and enjoy "playing doctor" by answering case studies. The podcast, *This Week in Parasitism*, provides real life examples of parasitic case studies. During lab, I play one case study and reward the correct answer with an extra point on their weekly quizzes. Students may use any resource and discuss possible answers among themselves. This low-pressure activity helps students piece together what they already know while motivating them to find answers to what they do not know. As they work through each case study, they learn more about the parasites they've been examining in the lab. Their last case study is intentionally difficult so students can generate a differential diagnosis. Second, I provide several ways for students to exercise their creative side by writing a parasite haiku, creating a parasite meme as well as (for graduating seniors) write, perform, and record a parasite song. Examples of their creative output will be provided.

6

Enhancing learning outcomes: specifications grading in parasitology

Kevin Garcia, Ana Elisa Garcia-Vedrenne
University of California, Irvine, Irvine, CA, USA.

Most educators agree that the worst part about teaching is grading. Traditional scoring has students and instructors debating over points and partial credit, rather than focusing on learning. Alternative grading formats like specifications grading (specs grading) are gaining attention for their potential to shift the focus from grading to learning, thereby improving student content knowledge and motivation. The specs grading approach aims to align course assessments with learning outcomes and provide transparency on how students can demonstrate achievement. Assignments are usually evaluated on a pass/fail basis and allow opportunities for reattempting or resubmitting assignments to demonstrate learning over time. In this study, we detail our implementation of specifications grading in an upper-division Parasitology class. The course targeted various learning goals that ranged from recognizing major parasitic groups and demonstrating knowledge of key disciplinary concepts to interpreting parasitological data and synthesizing primary literature in parasite ecology. With these learning goals in mind, we structured course components (e.g. quizzes, in-class activities, journal club presentations, project reports, and project presentations) into bundles tied to specific learning outcomes. Student feedback on the specs grading system was favorable, with many of them expressing their appreciation for the transparency and focus on growth over time.

7

Designing Effective Public Engagement in Parasitology

Hannah Bialic
University of Wisconsin, Madison, WI, USA.

Public engagement and science communication are integral to developing trust and understanding between the general public and research. It is especially relevant for the field of parasitology, as community engagement is fundamental to collecting patient samples and progressing treatment campaigns in endemic areas. This workshop will allow members of the symposium to engage with the current methodologies in place for designing effective public engagement, both domestically and internationally, and apply those methodologies to design novel engagement activities.

8

Network specificity decreases community stability and competition among avian haemosporidian parasites and their hosts

Daniela de Angeli Dutra[1,2], Robert Poulin[2]
[1]Stanford University, Stanford, CA, USA. [2]University of Otago, Dunedin, New Zealand.

Parasites play a fundamental role in shaping ecological communities and influencing trophic interactions. Understanding the factors that drive parasite impacts on community structure and stability (i.e., resilience to disturbances) is crucial for predicting disease dynamics and implementing effective conservation strategies. In this study, using avian malaria and malaria-like parasites as a model system, we investigated the relationship between specificity, community stability, and parasite vulnerability and their association with host diversity and climate. By compiling occurrence data from a global avian haemosporidian (malaria and malaria-like) parasite database (MalAvi), we constructed a comprehensive dataset encompassing 60 communities. We utilized a phylogenetic model approach to predict missing host-parasite interactions, enhancing the accuracy of our analyses. Network analyses based on bipartite interactions provided measures of network specificity, stability, modularity, parasite competition, and vulnerability to extinction. We found that the high network specificity reduced community stability and decreased competition among parasites. Furthermore, we found that parasite vulnerability decreased with increasing community stability, highlighting the importance of community stability in host-parasite interactions for long-term parasite persistence. When exploring the influence of local host diversity and climate conditions on host-parasite community stability, we demonstrated that host biodiversity and precipitation reduce parasite competition. Conversely, temperature raises competition among parasites. These findings provide valuable insights into the mechanisms underlying parasite impacts on communities and the interplay between specificity, community stability, and environmental factors. Further, we reveal the role of climate in shaping host-parasite interactions. By unravelling the complexities of parasite-mediated interactions, our research substantially improves the current knowledge of the importance of specificity as a modulator of interactions in bipartite networks.

9

Left high and dry: the interactions between trematodes and pond drying events in Boulder, Colorado

Sophie EM Elliott, Dana M Calhoun, Pieter TJ Johnson

University of Colorado Boulder, Boulder, CO, USA.

The frequency and intensity of drought conditions in the western United States have increased over the past several decades and are forecasted to worsen in the future. Wetlands and their aquatic inhabitants are highly vulnerable to drought, including the freshwater snails that function as important hosts for trematode parasites. During a survey from 2007-2009, trematode infections in pond snails were found to be rare in Boulder. However, in the summer of 2022, a high prevalence and diversity of trematode infections were detected in aquatic snails in South Boulder. This presented an opportunity to track the populations of these trematode taxa, and their snail hosts, over the course of a drying event to investigate how parasites respond to progressive dry downs in ponds. Using a natural experiment, the following questions were explored: a) how does drought impact snail density?, b) how does infection status and living status of snails change with time, and what impact does snail size have on those variables?, and c) how does the overall prevalence of each trematode taxon change with time (in the case of a perennial pond)? There were three ponds of focus (two ephemeral that dry out seasonally and one perennial that holds water year round), and biweekly field samples per pond from early September to mid-November 2022 were carried out. Dissections of nearly 3,000 snails of two species revealed the detection of six trematode taxa. Results indicated that snail density decreased significantly over time at the ephemeral ponds but increased at the perennial pond. Larger snails were disproportionately more likely to die based on the recovery of dead and empty shells at the ephemeral ponds. At the perennial pond, where ~20% of snails were infected, infection prevalence was greatest among larger snails and showed significant declines through time, although prevalence differed among trematode taxa. This is likely due to declines in the addition of new infections to ponds over time. These findings provide insight into how trematodes in Boulder could be impacted by future drought conditions and raise important questions about why infections in 2022 appeared to increase relative to earlier surveys.

10

Past vs. present: a survey of the fish parasites of the tributaries of Oneida Lake, New York

Hannah G Whitcomb, Florian Reyda
State University of New York at Oneonta, Oneonta, NY, USA.

Oneida Lake is the largest in-state lake in New York state and a premier fishing destination. In 1932, Van Cleave and Mueller completed a study of the parasite fauna of the fish in Oneida Lake and its tributaries, where they discovered and described thirty-three species of parasitic worms. Since 1932, the lake has undergone countless environmental changes which have impacted the invertebrate fauna, mollusks in particular. Recent (2016-2023) survey work was conducted on the fish parasites of the lake in order to compare to the results of Van Cleave and Mueller. The present study is a fish parasite survey of multiple tributaries of Oneida Lake to compare to both the recent and the past survey data of the lake itself. The methods of this study included the collection of thirty-three fish species via backpack shocking, e-boat and hook and line fishing, partial necropsy of fish to collect parasites and the mounting of parasites for identification using light and scanning microscopes. During the recent survey of the lake, certain species of trematodes that had been reported by Van Cleave and Mueller were considered 'missing.' Some of the missing species of parasites were found in the survey of the tributaries. For example, we encountered the trematode *Bunodera sacculata* in 3 of 31 *Perca flavescens*. *Bunodera sacculata* uses a native clam as its first intermediate host, which is thought to have been extirpated from the lake due to invasive species. In addition, the stream survey resulted in recovery of some species not originally reported by Van Cleave and Mueller. For example, eleven species of acanthocephalan were found, whereas only four were originally reported. The stream survey results also include multiple species of nematodes and cestodes, such as *Eustrongilides tubifex* and *Glaradacris catostomi.* This survey fills a

knowledge gap on the fish parasite data in the tributaries and provides context for the contemporary survey of the lake. Our survey demonstrates that the fish parasite species in the tributaries somewhat differ from those found in the lake.

11

Parasites of the Eastern Everglades, Florida fishes: Bangham 1940 revisited

Katerina D Sawickij[1], Christopher Blanar[2], J Matthew Hoch[2], David Kerstetter[2], Florian Reyda[1]
[1]State University of New York at Oneonta, Oneonta, NY, USA. [2]Nova Southeastern University, Davie, FL, USA.

We surveyed intestinal parasites of freshwater fish across the eastern portion of the Everglades, Florida from May to August 2023. The aim of this survey was to compare results to a classic parasite survey done in the area by Bangham in the late 1930's. We investigated 715 fishes of 31 species from 13 sites including storm water treatment areas, water conservation areas, Lake Okeechobee and surrounding water bodies. Fish were collected by seining, minnow trap, electrofishing, hook and line angling, and donation by local anglers. Intestines were examined for all fish and a subset received full necropsy. Only 15 of Bangham's original 45 species of fish were investigated in this study, reflecting the extensive environmental change in the habitat since the original study. Analysis is ongoing, but of the parasites observed, more immature worms were found than adult worms. Of the 15 overlapping host species between the two surveys, we only recovered a small subset of the original 22 adult parasite species. We documented several new locality and host records for parasites from native and invasive host species. From *Micropterus* sp. a new species of *Neoechinorhynchus* was documented with a unique egg morphology unlike any North American *Neoechinorhynchus* species. Also, from *Micropterus* sp. a poorly-known, previously described species, *Neoechinorhynchus doryphorus*, was documented. Three non-native species of parasites were documented, including the invasive Asian fish tapeworm, *Schyzocotyle acheilognathi*, from *Notemigonus crysoleucas*, a *Neoechinorhynchus* species from *Mayaheros uropthalmus*, and a trematode from *Channa marulius*.

12

The coccidia species *Pfeifferinella ellipsoides* exhibits spatial and species-specific variation in prevalence among freshwater snail hosts

Maria R Bollinger, Taylor M Fiedor, Kyle D Gustafson
Arkansas State University, Jonesboro, AR, USA.

There are fundamental data deficiencies related to our understanding of directly transmitted parasites, such as coccidia, for freshwater snails which are commonly studied for their role as intermediate hosts for digenetic trematodes. Coccidia likely play a role in structuring snail communities because of variation in pathogenicity and transmission among snail species. We collected freshwater snail species from the Ozark and Ouachita Mountains ecoregions in Arkansas to determine the spatial distribution and prevalence of coccidia in local freshwater snail species. We opportunistically collected 220 freshwater snails from 24 Ozark sites in summer 2022 and 146 snails from 19 Ouachita sites in summer 2023. We scanned snail fecal slides for coccidia. We scanned a subset of snails (*Physa* and *Planorbella* in the Ozarks) that did not have concurrent trematode infections and a subset that did to look for possible interactions among coccidia and trematodes. We observed oocysts that morphologically conformed to *Pfeifferinella ellipsoides* in 2 of the 9 snail taxa from 7 of the 43 sites. We observed *Pf. ellipsoides* oocysts in *Planorbella trivolvis* feces at 2 of 6 sites in the Ozarks and 0 of 5 sites in the Ouachitas. We found oocysts in feces of *Physa* species at 6 of 14 sites in the Ozarks and 0 of 12 sites in the Ouachitas. The overall prevalence was 0.13 (6 of 47) in Ozark *Pl. trivolvis* and 0.08 (8 of 97) for individuals in the genus

Physa. We ran chi-square and Fisher exact tests that revealed no significant evidence for trematode–coccidia competition or synergism within the two snail species. No other snail species was infected, and no *Pf. ellipsoides* oocysts were observed in snails from the Ouachitas. Our survey of 366 snails (9 taxa and 43 sites) represents the largest survey for freshwater snail coccidia to date. Our results indicate that both *Pl. trivolvis* and *Physa* sp. may be primary hosts and/or reservoir hosts for *Pf. ellipsoides* in freshwater snail communities. Further research is needed to investigate the highly aggregated distribution of *Pf. ellipsoides* in Arkansas. Our results led to several hypotheses for additional research, including questions regarding coccidia host specificity and virulence.

13

How does digenean trematode host specificity affect the biodiversity-disease relationship?

Brooke A McPhail, Patrick C Hanington
University of Alberta, Edmonton, AB, Canada.

There has been much debate surrounding the impact of biodiversity on disease transmission, with a special interest in digenean trematodes because of their complex life cycles. Additionally, the impact of biodiversity on host-parasite interactions is likely to be affected by whether the trematode community is comprised mainly of host-specialists or host-generalists. We hypothesize that digenean trematode species richness will relate positively with host biodiversity when host-generalists, rather than host-specialists predominantly define the trematode population. To test this hypothesis, snails were collected biweekly at 8 wetland sites to assess the trematode community during the months of May to August between 2019-2022. To assess overall biodiversity at the sites, field cameras, birdsong recorders, and benthic kick netting were also employed. Cercariae from each infected snail were identified using Cytochrome c oxidase 1 (*cox1*)-based DNA barcoding, and compared to our extensive database of *cox1* sequences representing over 90 unique trematode species. An additional summer of environmental DNA sampling was performed in 2023 for comparison to the snail data. Throughout this study, 22 396 snails were collected, of which 1 979 were infected with a digenean trematode (8.84%). Sixty trematode species belonging to 9 families have been identified. Based on supplemental biodiversity assessments, 136 species of vertebrates and 59 species of invertebrates have been identified from the sites. Network metrics will be used to study the effect of biodiversity on specialist and generalist trematodes. To date, most of the research that has categorized host specialists and generalist trematodes has focused on the definitive hosts, while this research will incorporate known snail hosts. By combining our extensive database of trematodes known to be present in Alberta with the biodiversity data collected during this reclaimed wetland study, we can address the much-debated question surrounding the relationship between biodiversity and disease.

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Parasitic Varroa Mites Employ Kleptoparasitism on the Molecular Level to Accelerate Reproduction

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Rapid reproduction is among the most significant adaptations underpinning the success of the honey bee parasite *Varroa destructor* (Class: Acari, Order: Mesostigmata). To exploit their host, the parasitic mite must produce offspring that fully develop into adults and mate in an inflexible nine-day window. Failure to meet this deadline brings the foundress mite's fitness to zero establishing heavy evolutionary pressure to accelerate reproduction and subsequent development of offspring. Through dynamic energy

budget modeling, we've assessed the energy invested in reproduction to be more than an order of magnitude greater than the expected input. Such rapid production of egg yolk proteins would seem to outstrip the capacity of the parasite's ribosomes. A biological feat of this nature requires explanation. Via Native-PAGE, we confirmed that nine proteins detected in honey bee fat body tissue accumulate intact in the foundress mite’s eggs. Using antibodies raised against honey bee vitellogenin, we identified this egg yolk precursor via SDS-PAGE and concomitant immunodetection. We then analyzed samples of honey bee fat body and *Varroa eggs* via HPLC MS/MS to identify the remaining host proteins. We detected egg yolk precursors in the large lipid transfer protein superfamily, in addition to hexamerin storage proteins, and miscellaneous motor/transfer proteins integral to embryonic development. *Varroa* cannot produce some of these proteins and instead employ kleptoparasitism on a molecular level to provision their developing ova, a pathway not described in any other host/parasite relationship. We have named this process kleptocytosis. Better understanding of this pathway presents an attractive target for *Varroa* management as a treatment would only need to delay development of the mite to render reproduction unsuccessful.

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Canine Hookworm Presence in Tongatapu, Tonga

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Dogs are common in Tonga, where they exist in a close relationship with humans as pets and guard dogs and often are free-roaming due to limited property fencing and the need to scavenge for food. Little is known about the health status of these dogs and the type and level of pathogens carried by them. In this study, blood and fecal samples were analyzed to identify the presence of endoparasites and determine packed cell volume (PCV) values for dogs in Tonga. The current data deficit in this area made it necessary to develop a methodology and collect preliminary data to assist with future research in Tonga. Fecal analysis revealed hookworm in 73% of dogs, primarily those 4–12 months old. Hematologic evaluation revealed low to borderline low PCV in 87% of dogs. The zoonotic nature of some of the pathogens identified poses a public health risk, with a particular risk for children.

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Regulation of sexual differentiation in mantids: A novel perspective on the impact of hairworm on host development

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The hydrotactic behavior observed in hairworm-infected insects is evidently advantageous for the parasite and detrimental to the host. Serving as an extended phenotype with adaptations for both the host and parasite, the hairworm manipulation demonstrates a pronounced bias toward the parasite. An influential factor driving this profound manipulation is the early castration induced by hairworm infection, leading the host to forfeit the opportunity to pass on its genes, resulting in evolutionary dead ends. Infected mantids display various traits signaling reproductive reduction, such as reduced egg production in the ovaries of female adults and gonadal loss in male adults, accompanied by intersexual characteristics like a diminished antennal sex pheromone receptor and feminized wing shapes. Hairworms actively interfere with sexual differentiation during the early stages of infection, with intersexual characteristics in male antennae becoming apparent within one month of exposure to hairworm cysts. Tracking the developmental trajectory of infected mantids suggests that the emergence

of intersexual characteristics goes beyond a mere outcome of parasitic juvenilization, which halts male development before sexual differentiation. Instead, infected males appear directed toward a female developmental trajectory. In parallel with hairworm infection, the presence of males with feminized wings is observed in the *Amantis nawai* population. Within this population, males exhibit two wing types, long-winged morph and short-winged morph, while females exclusively display the short-winged morph. Insights derived from hairworm infection suggest the potential applicability of a similar scenario to mantids exhibiting wing dimorphism, a phenomenon that might have been overlooked.

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Behavioural and proteomic analysis of the role of dehydration in crickets, *Acheta domesticus*, infected with the nematomorph *Paragordius varius*

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Nematomorphs are a group of parasitic worms which infect arthropods as their definitive host. To complete their lifecycle, these worms manipulate host behaviour, resulting in water entry by the host. The mechanism influencing this behaviour is unknown, though light attraction, water attraction and erratic behaviour have been implicated. Here we investigated the attraction to water hypothesis using crickets (*Acheta domesticus*) exposed to the nematomorph *Paragordius varius*, with the aim of determining if water-seeking behaviour is driven by similar mechanisms to dehydration. A 'Y' maze choice test with a water trough at the end of one arm and an empty trough at the end of the other was used to assess the water attraction behaviour of crickets. As expected, crickets infected with a nematomorph were more attracted to water than uninfected crickets. Post-infected crickets, allowed to expel their worms prior to testing, favoured the dry trough, indicating that water-seeking behaviour is reversible once the parasite has been expelled. Uninfected dehydrated crickets were more attracted to water than control crickets but not to the extent of infected crickets. Dehydrated crickets did not fully enter the water, as the infected group did, but instead submerged their heads under the water for a prolonged period of time, indicating a level of consciousness of the danger of the water. Time of day had no effect on water-seeking behaviour. We have used mass spectrometry to establish a proteomic signature of dehydration in cricket haemolymph and compared this to the proteomic signature of infected crickets haemolymph. To conclude, crickets infected with a nematomorph are more likely to enter water than control crickets and this behaviour is reversible. Dehydration may be partially involved in driving water-seeking behaviour, however, this does not fully account for the water-seeking behaviour observed in infected crickets, suggesting other mechanisms are also involved.

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Exploring the sensory landscape of hairworm-infected crickets

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Many parasites have the adaptive ability to modify the phenotype of their host to complete their life cycle, a trait known as host manipulation. Since environmental cues trigger the rapid cascade of biochemical interactions that elicit behavioral responses in animals, we expect to see differences in the responses of infected animals toward these cues, especially if a particular cue or set of cues favors parasite transmission or development. For example, if a crustacean infected with a trematode responds less to predator cues, this may decrease escape behaviors, rendering the crustacean more vulnerable to predation and favoring the development of the trematode via trophic transmission. To understand these complex interactions with external stimuli, we must test cues in isolation, to better map the sensory landscape guiding behavioral manipulation. In freshwater hairworms (Nematomorpha: Gordiida), mature individuals somehow "drive" their terrestrial insect hosts into water, where the insect can die, and the hairworm exits to mate. Infected insects are likely more active at night and respond to environmental cues differently than uninfected ones, however, we do not know how these differences evolve throughout hairworm development. We hypothesize that the modified responses to environmental stimuli appear only during the critical period preceding hairworm egression. Using *Acheta domesticus* crickets experimentally infected with *Paragordius varius* hairworms in a time series of behavioral assays, we tested multiple stimuli to determine the extent to which hairworms modulate host behavior, and whether these behaviors correlate with hairworm development. Humidity, horizontally polarized light, and direct contact with water were tested in isolated behavioral assays, all of which were recorded under high-definition cameras and infrared light. Among other observations, we noticed a remarkable behavior reminiscent of apple bobbing in the water assay, where the crickets would repeatedly dunk their heads into water. Also, hairworms apparently needed to contact water to egress, otherwise they would retreat into the abdomen of their host. We discuss these behaviors in tandem with the responses of infected crickets to humidity and light, and link these to the sensory landscape in which these remarkable behaviors are observed.

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Bacterial resistance and longevity of crickets infected with Gordian worms (Nematomorpha)

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The presence of parasites induces an acute stress response in hosts, altering their physiological state. This stress response is often associated with immunosuppression leading to increased susceptibility to bacterial and viral infections. This eco-immunological trade-off has been extensively studied in arthropods, including crickets. The Gordian worm *Paragordius varius* depends on its cricket host, *Acheta domesticus*, for resources to grow from tiny larvae to adults reaching 25cm in length and often having the same body mass as the host. At the culmination of the host-parasite association, the parasite relies on a physiologically-capable host for transport and release into aquatic environments, where *P. varius* reproduces. This symbiotic relationship presents a dilemma, as the need for a healthy, immune-competent host conflicts with the immunosuppressive effects induced by the significant stress of hairworm infection, affecting the energy balance within the host-parasite unit. To investigate this conflict, we conducted experiments assessing the resistance of the host-parasite unit to tertiary bacterial threats and its long-term survival. Adult female crickets were exposed to 10-50 *P. varius* cysts, while controls underwent sham exposure. At 21 days post-exposure (DPE), crickets were injected with an LD50 dose of the bacterial insect pathogen *Serratia marcescens*. Mortality in both groups was monitored until 31 DPE, after which crickets in the experimental group were dissected to confirm infection with *P. varius*, with uninfected individuals removed from further analysis. In a subsequent experiment, we compared the survival rates of *P. varius*-infected crickets with those of uninfected controls. Surprisingly, findings revealed that the experimental group, also infected with *P. varius*, exhibited greater survival success against LD50 doses of *S. marcescens* compared to uninfected control crickets. Furthermore, survival curves indicated greater short-term and long-term survival of *P. varius*-infected crickets compared to control crickets. These results suggest that Gordian worm infections may enhance immune activity in host-parasite units, ultimately leading to greater disease resistance and

increased longevity compared to uninfected control animals. Although the specific mechanisms underlying these results remain undetermined, several alternative explanations will be discussed.

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Hairworm abundance and prevalence as an indicator of ecosystem health

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As human activity continues to threaten natural ecosystems, the need for a cheap, quick, and reliable method to assess ecosystem health increases. Parasites with complicated life cycles such as hairworms [Nematomorpha] may provide a solution. Hairworms infect various terrestrial arthropods, which they manipulate into entering water where the adult worms emerge, mate and lay eggs. Eggs hatch into larvae which encyst in a second arthropod host, usually an aquatic insect larva which, when it becomes a flying adult, moves the hairworm to the terrestrial environment. Because hairworms require at least two arthropod hosts to complete their life cycle, the absence of either host could result in the parasite becoming locally extinct. Thus, we hypothesize that hairworm cysts will be found at a significantly higher prevalence in more pristine water and where aquatic insect diversity is higher. To test this, we sampled 50 sites near Lincoln, NE for aquatic snails and insects and measured water parameters (nitrate, DO, pH, salinity, and atrazine levels). These sites were previously sampled (Hanelt 2001) for hairworm cyst prevalence and abundance as measured by counting hairworm cysts in 20 *Physa* aquatic snails. Thus, we further hypothesize that hairworm cyst prevalence and abundance will remain unchanged compared to their prevalence and abundance in 2001 except at sites that experienced human disturbance. Four of the 50 sites tested positive for atrazine (above 3 ppb). Insect diversity, measured with the Shannon Diversity index, will be correlated with hairworm cyst prevalence and abundance. Results are discussed in the context of using hairworms as indicators of aquatic ecosystem health.

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The hairworm and the cricket: a story of life and death

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The phylum Nematomorpha is an obscure group of meiofauna that live in freshwater and marine ecosystems. Freshwater Gordiids have complex life cycles that incorporate both aquatic and terrestrial invertebrates. To date, 370 freshwater species have been described but it has been suggested there are more than 2000 species. In the last 20 years numerous studies of biodiversity, host use, and distribution have been documented. However, few studies have examined how these parasites affect the development of hosts. Often described as parasitoids, a recent study showed female cricket hosts can survive infection. But studies on the hairworm's effect on males are lacking. In this study, we examined the effects of hairworm development on both male and female laboratory reared crickets to determine the mortality rate development rate (body length, mass, wing length), and lifespan to determine costs exerted by hairworms on their hosts. To infect crickets, freshwater pond snails were exposed to laboratory-reared hairworm larvae. After 2-3 weeks, larvae became encysted in snails and were then fed to two-day starved crickets. Body length, mass, and wing length were recorded upon onset of the experiment, repeated when the worms emerged, and at the date of cricket death. We also examined correlations between host size and parasite size. The significance of development and an examination of a parasitoid lifestyle will be discussed.

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Life History Diversity of the Freshwater Gordiids (Phylum: Nematomorpha).

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The phylum Nematomorpha consists of two taxa, the freshwater Gordiida and the marine Nectonematida, and represents one of three entirely parasitic animal phyla. Gordiids have complex life cycles which include multiple hosts and a free-living aquatic phase. The typical life cycles reported for Gordiids includes a parasitic phase, where Gordiids manipulate the behavior of their terrestrial arthropod hosts to enter aquatic environments where adult worms emerge. After emerging from their host, male and female worms form Gordian knots, mate, and females deposit egg strings as free-living forms. Females produce millions of eggs during their short (2 weeks–2 months) free-living phase. Within weeks, larvae develop, infect, and encyst indiscriminately within a variety of aquatic vertebrate and invertebrate animals. Some of these aquatic animals (insects) act as paratenic (transport) hosts by carrying cysts to land where they are consumed by omnivorous or predatory definitive hosts including millipedes, orthopterans (crickets, grasshoppers, etc.), beetles, cockroaches, and mantids. However, and over the last 20 years, our work on the life cycles of freshwater Gordiids has revealed species that are parthenogenetic, strictly terrestrial, and strictly aquatic in their life cycles. In this presentation we will review how common the other Gordiid life history strategies are, and we will provide information on field and laboratory techniques useful for documenting life cycle diversity among the Gordiida, to better understand their global diversity and life histories.

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Biodiversity of horsehair worms (Nematomorpha): advances and challenges

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New species of horsehair worms (Nematomorpha) are constantly discovered and described. 77 of the known 370 species were described in the past 25 years. The new descriptions extend the biogeographic range of genera. This is most evident in the genus *Beatogordius*, for which species were originally described from South America and Africa, but were later discovered in Australia, India and China. The latest new genus was described in 2000. Most new species are added in the already species-rich genera *Chordodes* and *Gordius*. Still most species are described on the basis of single or very few specimens, which is valid, but not uncritical. There are examples for the existence of cryptic species and, at least in some species, strong phenotypic plasticity appears likely. Molecular barcoding is still in its infancy in nematomorphs, but may become a strong and helpful tool.

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Bma-LAD-2 as a possible vaccine target against *Brugia malayi*

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Efforts to reduce the global burden of Lymphatic Filariasis focus primarily on Mass Drug Administration (MDA). While highly effective against microfilariae, current MDA regimens have little to modest effects on adult worms. Thus, MDA typically has to be repeated over several years. Additionally, the most efficacious MDA regimen is contraindicated in areas co-endemic with *Loa loa* and *Onchocerca volvulus* infections due to adverse effects caused by rapid killing of microfilariae. The long-term goal of this study is to develop a novel macrofilaricidal agent that would allow for fewer MDA treatments and

safe use in areas with *L. loa* and *O. volvulus*. Previously, we showed that Bma-LAD-2, an intestinal protein in *Brugia malayi* likely involved in tight junction formation, is essential for adult worm fecundity, metabolism, and motility. In this study, we are testing whether antibodies targeting Bma-LAD-2 can function as macrofilaricidal agents. We have recombinantly expressed a soluble form of the outer binding Ig domains of Bma-LAD-2 (Bma-LAD-2 Ig1-4) and have created stably transfected HEK293 cells expressing full-length transmembrane Bma-LAD-2 protein. Bma-LAD-2 transfected cells were found to have higher electrical resistance across a monolayer than non-transfected HEK293 cells by TransEpithelial Electrical Resistance (TEER) assay. Addition of polyclonal rabbit antibodies against Bma-LAD-2 Ig1-4 to the transfected cell line decreased TEER values to those of non-transfected cells. Finally, we have found that anti-SARS-CoV-2 antibodies in human sera cultured with adult *B. malayi* worms were still functional after ingestion by adult worms. Together, these results suggest that adult filarial worms ingest antibodies into their intestinal tracts, and that antibodies against Bma-LAD-2 have the potential to interrupt intestinal tight junctions of filariae. We are currently working to determine the effect of treating adult *B. malayi* worms with anti-Bma-LAD-2 antibody. These findings suggest that targeting nematode intestinal tract integrity may be a promising avenue for development of novel anthelmintics.

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Peripherally circulating immune cell profiles in African children indicate a heterogeneous immune response against *Plasmodium falciparum* malaria

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Malaria remains a global health challenge with over 600,000 deaths annually despite 70 years of worldwide elimination efforts. Ninety-five percent of the malaria burden comes from sub-Saharan Africa, although autochthonous malaria has re-emerged in Florida and Texas. With a long history of co-evolution between humans and *Plasmodium* species, many immune evasion techniques are employed by the parasite that have challenged vaccine development. In regions with moderate to high transmission rates, children are repeatedly infected and eventually develop naturally acquired immunity (NAI) that is characterized by protection against malaria illness but not complete elimination of blood-stage infection. This phenomenon promotes undetected asymptomatic infections that are the largest contributor to the ongoing transmission of malaria. The mechanisms that differentiate NAI and sterilizing immunity are poorly understood. We analyzed cellular proportions in children who are developing NAI at an asymptomatic baseline, during uncomplicated *Plasmodium falciparum* malaria, and upon recovery in whole blood of 57 children from Burkina Faso and in peripheral blood mononuclear cells of 8 children from Kenya. We obtained cell proportion estimates from reference-based cell-type deconvolution strategies with both RNA sequencing and paired genome-wide DNA methylation data. We discovered multiple patterns in cell type proportion shifts during clinical malaria relative to their baseline and recovery estimates using principal components. K-means clustering identified at least 2 groups in children from Burkina Faso and 2 in the children from Kenya. The proportions of monocytes, naive CD4 T cells, and other cell types differ by cluster in a pattern that is consistent across the two populations. Furthermore, preliminary results revealed unique gene expression and DNA methylation profiles during malaria and at recovery when stratified by the k-means identified clusters. The etiology and downstream susceptibility to future infections for this heterogeneous immune response is yet to be understood but does not relate to age or sex. These ongoing analyses question the commonly held assumption that all children have the same immune response during an uncomplicated malaria illness. The existence of multiple responses can substantially change how immune studies are designed, analyzed and interpreted.

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Schistosomiasis: Principal requirements for the development of a molecularly defined vaccine

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Schistosomiasis is a debilitating neglected tropical disease caused by infection with parasitic trematodes of the genus *Schistosoma*. Six species of *Schistosoma* are the basis of clinical disease in humans, altogether responsible for over 290,000 deaths annually. The clinical manifestations of schistosomiasis are chronic and insidious, including fever, anemia, growth retardation, genital lesions, hepatosplenomegaly and irreversible organ damage. According to recent estimates, schistosomiasis is endemic in 78 countries with over 800 million people at risk for infection. Despite mass drug administration programs with praziquantel, the prevalence of schistosomiasis remains high. A vaccine is urgently needed to control transmission of this disease. Making a vaccine for a multicellular eukaryotic parasite whose genetic machinery is exactly similar to their host, requires more detailed understanding of both host as well as the parasites. An effective schistosome vaccine could offer sustainable reduction in both the prevalence and transmission of the disease. In May 2022, Infectious Diseases Clinical Research Consortium (IDCRC) researchers started conducting a clinical trial of Sm-p80/GLA-SE (SchistoShield). The trial could help pave the way for the world's first vaccine against the disease, offering a safe and cost-effective option to lower its prevalence worldwide. Before the human clinical trials of Sm-p80/GLA-SE (SchistoShield), this investigational vaccine had been extensively tested in more than three hundred baboons to qualify for the Phase-I clinical trials. The main reason for this requirement is that while working to develop a molecularly defined vaccine for a multicellular eukaryotic pathogen, most of the time, mice data cannot be replicated in non-human primates. A summary of the fundamental requirements for the development of a subunit vaccine will be discussed at the conference.

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Toll-like receptors in snail hemocytes and beyond

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The snail, *Biomphalaria glabrata*, is an emerging model organism since it is the obligate intermediate host for *Schistosoma mansoni*, a trematode parasite impacting millions of people annually. Since mass drug campaigns targeting *S. mansoni* in humans have proven ineffective for parasite population control, we aim to better understand the host-parasite interactions occurring within the snail. Although the field of gastropod immunology is relatively sparse, several immune factors have been functionally characterized for their role in coordinating snail resistance to worm infections. However, it remains unclear where the main anti-schistosome effector cells come from both with regards to their spatial origin and the cell lineage from which they differentiate. Here, we present data collected by flow

cytometry to show that the reactive oxygen species used for parasite killing are predominantly generated by granulocytic cells expressing a unique toll-like receptor (TLR). Additionally, the hematopoietic boundaries between hemocyte populations have been further characterized using proliferation assays in the context of growth factor stimulation. To address immune dynamics in non-hemocyte tissues, we have developed methods to see how molecular signaling directs parasite encapsulation in vivo. Somewhat unexpectedly, we found TLR expression not only in the encapsulating hemocytes but also in visceral tissues which have, to this point, been overlooked in snail immunology. Our results suggest that there are multiple pathways to TLR+ granulocyte differentiation, including from circulating cells and proliferative hematopoietic sites. Thus, our findings set an important precedent for looking beyond hemocyte cells to fully understand functional resistance to *S. mansoni.*

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The extracellular matrix proteins Pericardin and Lonely heart mediate periostial hemocyte aggregation in the mosquito *Anopheles gambiae*

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The immune and circulatory system of a mosquito are functionally integrated, which allows for a dynamic response to pathogenic invasion. Following infection, the primary immune cell of the mosquito, known as the hemocyte, aggregates to the mosquito heart specifically along valves called ostia. At the ostia, hemocytes experience a higher rate of hemolymph flow which maximizes the chance they encounter a pathogen. Furthermore, the hemocytes that are aggregated around the ostia, called periostial hemocytes, appear to modulate how hemolymph is circulated during an infection—decreasing the heart rate and skewing the proportional directionality of heart contractions. Despite the importance of periostial hemocytes on both circulatory and immune physiology, the molecular mechanism that drive hemocytes to aggregate on the heart are not well understood. Here, we investigated the role of the extracellular matrix proteins, pericardin and lonely heart, in the periostial hemocyte aggregation response of *Anopheles gambiae* to infection with *E. coli*. We found that silencing pericardin or lonely heart via RNAi decreases the mean number of periostial hemocytes by 59% and 61%, respectively, following *E. coli* infection. Furthermore, preliminary data suggest that periostial hemocytes are necessary for infection-induced changes in circulatory physiology; when hemocytes are removed via chemical ablation, bacterial infection neither decelerates the heart rate nor changes the proportional directionality of heart contractions. Altogether, the extracellular matrix proteins, pericardin and lonely heart, facilitate the aggregation of hemocytes on the heart, and periostial hemocytes appear to be required for infection-induced changes to circulatory physiology.

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*Pf*ATAD3 is a parasite-encoded protein critical for mitochondrial biogenesis in *Plasmodium falciparum*

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Malaria is an infectious disease that has plagued humanity for more than 500,000 years. *Plasmodium falciparum,* specifically, accounts for about 95% of malaria-attributed deaths. Lately, there has been an exponential rise in the rates of malaria-related deaths due to antimalarial drug resistance, insecticide resistance, and global warming. Consequently, we must continue to expand our toolkit against malaria by studying parasite physiology and gaining more insight into plasmodial proteins that can serve as potential antimalarial drug targets. Although the *P. falciparum* mitochondrion is one of the validated antimalarial drug targets, many *P. falciparum* mitochondrial proteins remain uncharacterized. This

project aims to define the roles of a newly identified mitochondrial *P. falciparum* protein, PF3D7_0707400, in mitochondrial biogenesis and parasite development. Bioinformatic analyses revealed PF3D7_0707400 to be an ortholog of ATAD3A (ATPase associated with diverse cellular activities), hence we refer to it as *Pf*ATAD3. To determine its essentiality in *P. falciparum* parasites, we employed the TetR-DOZI CRISPR-mediated inducible genetic knockdown approach. We explored the roles of *Pf*ATAD3 in mitochondrial biogenesis through live cell scanning confocal imaging of TOM22-mNG (Translocator of Outer Mitochondrial Membrane 22 - mNeonGreen) tagged *P. falciparum* parasites stained with MitoTracker, a membrane potential-dependent fluorescent dye. Additionally, we conducted a proteinase K assay and a large-pore agarose and polyacrylamide composite gel electrophoresis to elucidate the topology and native state of *Pf*ATAD3. Our data show that *Pf*ATAD3 is essential to parasite development as parasites die when *Pf*ATAD3 is not expressed for at least 96 hours. We observe a loss of mitochondrial membrane potential in about 60% of the parasites not expressing *Pf*ATAD3 for 48 hours indicating a role of *Pf*ATAD3 in maintaining mitochondrial membrane potential in asexual *P. falciparum* parasites. The results of the proteinase K assay and the composite gel electrophoresis demonstrate that *Pf*ATAD3 exists in a ~ 4 mega-Dalton complex spanning both inner and outer mitochondrial membranes with its C-terminus in the mitochondrial matrix and some of its N-terminus exposed to the cytosol. In conclusion, *Pf*ATAD3 is a mitochondrial protein that plays an essential role in mitochondrial biogenesis and development of asexual *P. falciparum* parasites.

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Extreme diversity of mitochondrial genome organization within the coccidian suborder Adeleorina

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Parasites in the coccidian suborder Adeleorina (Apicomplexa; Eucoccidiorida) are united by the association of macro- and micro- gamonts prior to gamete formation in a process called syzygy. Some of the most poorly classified and understudied apicomplexan parasites reside within this suborder. Most heteroxenous species have been classified based on stages visible in vertebrate intermediate hosts with many having poorly defined or unknown definitive hosts. Monoxenous species parasitize invertebrates with one exception, *Klossiella* species. Adeleorinid parasites typically have short (~6,000 bp) circular-mapping mitochondrial genomes encoding 3 protein coding regions (COI, COIII, CytB) and fragmented large and small subunit rDNAs. The organization and content of mitochondrial genomes from eimeriid coccidia are highly conserved among many genera; this has not been explored sufficiently in the adeleorinid coccidia. Publicly available mitochondrial genomes from adeleorinid parasites include 4 monoxenous species (3 *Klossia* spp. and *Klossiella equi*) and 10 from heteroxenous *Hepatozoon* species. Even among these few complete sequences, there is unexpected diversity in mt genome organization between the monoxenous families Adeleidae and Klossiellidae. Three recently generated *Haemogregarina* species mt genomes have conserved organization and contents with minor variations in the lengths of a few rDNA fragments. Monoxenous *Klossia* species were most similar in mitochondrial genome arrangement to heteroxenous *Haemogregarina* species rather than monoxenous *Klossiella* species. Despite their similar organization, the mitochondrial genome sequences of *Haemogregarina* and *Klossia* were less similar to one another than *Klossia* species were to *Klossiella equi*. Definitive host-parasite coevolution has been observed repeatedly in the phylum Apicomplexa suggesting that parasites with closely related definitive hosts might have more similar mitochondrial genomes (sequence and organization). The wildly divergent mitochondrial genome organizations observed within a single genus (i.e. *Hepatozoon*) may reflect their wide range of evolutionarily distant definitive hosts (e.g., insects, ticks, mites) that diverged between 635 and 538.8 million years ago. Similarly, genera within the Adeleidae likely have varied mitochondrial genome arrangements because of the evolutionarily distant phyla of their definitive hosts (tubificid worms, snails, insects). However, until more sequence data are generated, this and the true relationship of these parasites to the rest of the Apicomplexa remains unresolved.

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Mitochondrial genomes of coccidia with morphologically distinct sporocyst excystation structures

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The mitochondrial (mt) genomes of coccidia belonging to the suborder Eimeriorina are typically small (~6kb) and circular-mapping (but linear concatemers physically) that encode three proteins (CytB, COI and COIII) and a variety of fragmented rDNAs. The protein-coding regions are particularly well suited for species-level identification. The mt genomes of coccidia infecting more economically relevant homeotherms (e.g., *Eimeria* and *Isospora* species) are highly conserved with respect to gene content and organization. Organization and content of mt genomes from coccidia infecting fish, reptiles, or amphibia have remained largely unknown because these parasites of poikilotherms are examined infrequently in comparison to their homeotherm-infecting counterparts. This study was conducted to compare the mt genomes of members of the family Barrouxiidae, Léger 1911 typically infecting poikilotherms that possess sporocyst sutures for excystation, rather than Stieda bodies typical of coccidia in the family Eimeriidae. Complete mt genomes of three coccidia (one *Goussia* sp. and two *Choleoeimeria* spp.) belonging to the family Barrouxiidae were PCR-amplified, sequenced, assembled, and annotated to permit comparison with mt genomes from eimeriid coccidia infecting economically relevant homeotherms. The mt genome of *Goussia degiustii* (Molnar and Fernando, 1973) shared both content and organization with the mt genomes of species of *Eimeria*, *Isospora* or related eimeriid coccidia. In contrast, the mt genomes of the two newly sequenced *Choleoeimeria* species possessed the same gene content as their eimeriid relatives but with altered organization. A pair of previously reported inverted regions found in the mt genome of *Choleoeimeria taggarti* (i.e., LSUA inverted - 176bp; run of 6 rDNA fragments inverted - 729bp) were also present in the newly obtained genome sequences. All *Choleoeimeria* species examined, despite infecting distantly related hosts on separate continents, shared the same mt genome content and organization distinct from both *Eimeria* and *Goussia* species. These findings are only a glimpse into the variation of mt genome organization of coccidia parasitizing poikilotherms that has just begun to be explored.

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Methods for generating genomes of protozoan parasites with food safety importance directly from fecal isolates

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Pathogenic intestinal protozoan parasites that infect humans and other animals can be challenging to track and identify, which hampers epidemiological investigations into infection sources and the role of genetic diversity in pathogenicity. Whole genome sequencing and comparative genomics can assist in the elucidation of the traits that influence transmission and disease outcomes. However, culture methods to obtain sufficient DNA for whole genome sequencing is not feasible for most protozoan parasites important in food safety. Because of these challenges in DNA acquisition, genomic data from fecal isolates obtained directly from infected hosts are lacking and limit comparative genomics studies. To fill this data gap, we have developed protocols suitable for generating genomes from freshly isolated parasites from fecal samples and applied these methods to sequence and study genomes from multiple

animal hosts. We have tested and applied a variety of cleaning, concentrating, sequencing, and assembly techniques to isolates of *Giardia, Cryptosporidium,* and *Cyclospora* to produce genomes for these important human pathogens. By leveraging new sequencing and assembly strategies, we can begin to generate the data needed to understand the genetic basis of host specificity, pathogenicity, and transmission dynamics of these important human and animal parasites.

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Application of next generation amplicon sequencing to characterize genetic diversity of protozoan parasites of food safety importance

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Cryptosporidium and *Giardia* are enteric protozoan parasites that infect humans and many other animals. They are a significant cause of diarrhea worldwide and are responsible for numerous waterborne and foodborne outbreaks of diseases. Both parasites have species/genotypes that are morphologically indistinguishable but that display genetic variability and present different degrees of host specificity. Not all genetic variants cause disease or are human pathogens, making it crucial to correctly identify species/genotypes. Molecular methods are required to classify species/genotypes of these parasites, which currently rely on Sanger sequencing that lacks the ability to detect mixed species/genotypes within a sample. Therefore, mixed infections have been systematically underreported, and their role in transmission of zoonotic species/genotypes has not been fully explored. To address this problem, we have developed next generation amplicon sequencing (NGS) protocols and analysis pipelines to overcome Sanger sequencing limitations. We have demonstrated that NGS protocols are well-suited to detect mixed species/genotypes infections, low abundance genetic variants, and intra-species/genotype variants, which would have been missed using direct Sanger sequencing alone. NGS protocols are powerful tools for exploring *Giardia* and *Cryptosporidium* genetic diversity in infected hosts and environmental samples which may aid in understanding *Giardia* and *Cryptosporidium* epidemiology and zoonotic transmission.

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Characterizing genetic diversity and population structure of human hookworms using whole genome data from accessible sample types

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Human hookworms infect >400 millions of individuals worldwide and targeted treatment through Mass Drug Administration (MDA) of anthelmintic drugs reaches 100s of millions of people every year. Understanding how parasite populations are structured, and how that structure changes through time and in light of treatment campaigns, can provide an additional an important data source to measure the consequences of global health programs. Larvae are the ideal sample type for hookworm genomic studies as they are both more accessible from natural populations than adults and easier to manipulate than eggs, however the amount of gDNA extracted from individual larvae is insufficient for whole genome sequencing. In this study, we explore the use of whole genome amplification (WGA) of L3 larvae to generate complete genomic data from individual specimens. To first ensure that WGA does not systematically bias next-generation sequencing (NGS) datasets, we validated our approach using adult worms. Genomic DNA (gDNA) extracted from individual adults was serially diluted tenfold (10^{-1} to 10^{-4})

and dilutions were amplified using a GenomiPhi V3 Ready-To-Go DNA Amplification Kit. Amplified and unamplified whole gDNA was then sequenced on an Illumina NovaSeq 6000 targeting 50× depth of coverage per sample. Quality-controlled NGS data were aligned to a reference genome and single nucleotide polymorphisms (SNPs) were called from each sample using the GATK bioinformatic pipeline. GATK was then used to measure false discovery rate and genotype concordance between amplified dilutions and unamplified gDNA sequenced from the same individual to confirm that WGA does not introduce genotyping errors. Following validation, gDNA was extracted from individual L3 and the presence of gDNA was confirmed with qPCR. Successful extractions were used as input for WGA and sequencing following the methods described above. To assess genetic differences between individuals, SNP datasets generated from quality-controlled read data using GATK were analyzed using discriminant analysis of principal components (DAPC), STRUCTURE, and RAxML. Moving forward, this approach can be used to characterize hookworm population structure and diversity in natural populations.

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Loss of phenotypic traits in hookworms associated with recovery from cryopreservation

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Cryopreservation of parasitic nematodes is routinely used to avoid maintaining isolates in expensive animal models, as well as to provide a reserve that can be reanimated if an isolate is accidentally lost or contaminated. There are several methods used to cryopreserve hookworms either as infective third stage larvae (iL3), which can be used directly after thawing to infect a host, or as free-living first stage larvae (L1), which must be grown to iL3 on agar plates following recovery. Recently, a contamination event during life-cycle maintenance resulted in the mixing of a multi-anthelmintic drug resistant (MADR) isolate of *Ancylostoma caninum* (KGR) with our drug susceptible wild-type isolate WMD. To re-establish pure infections, motile iL3 recovered from stocks of both isolates that had been cryopreserved for over 1 year were used to infect naïve beagles. Upon characterization of the recovered KGR isolate (KGR^{rec}), we found that the isolate retained resistance to thiabendazole (TBZ) and ivermectin (IVM), but lost resistance to pyrantel pamoate (PYR). Furthermore, in the original KGR (KGR^{ori}) isolate, *in vitro* L3 activation was depressed approximately 40% compared to the wild type WMD isolate, whereas KGR^{rec} larvae activated to similar levels as the control WMD larvae. Our results indicate that long term cryopreserved (> 1 year) *A. caninum* iL3 can be recovered and successfully used to infect dogs, but that recovery from cryopreservation can result in loss of phenotypic traits in the recovered worms and subsequent generations. The loss of phenotypic traits following recovery likely results from genetic drift during passage through the extreme genetic bottleneck associated with recovery from cryopreservation.

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Investigating the anthelmintic potential of *Momordica charantia* against hookworm infections

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Hookworm infections pose a significant health threat in developing countries, particularly affecting vulnerable populations with limited healthcare access. The inevitable emergence of anthelmintic resistance, driven by widespread prophylactic drug administration underscores the urgency for novel treatment approaches. *Momordica charantia*, or bitter melon, is cultured widely in tropical regions where hookworm is endemic, and is often used in traditional medicine practice to treat intestinal parasite infections. We used two *in vitro* assays to investigate anthelmintic activity in *M. charantia*

extract (MCE). First, an activation assay was used with *Ancylostoma caninum* L3 larvae to evaluate larval feeding inhibition, examining effects of storage conditions on activity, impact on wild-type and resistant nematode isolates, and activity retention following organic extraction in various solvents. Second, a survival assay was used in which adult *Ancylostoma ceylanicum* are exposed to varying concentrations of MCE and lettuce (*Lactuca sativa*) extract, our control plant material, over 48 hours. Noteworthy findings include enhanced inhibition of larval feeding with prolonged frozen extract storage, heightened efficacy of *M. charantia* on anthelmintic-resistant *A. caninum*, and optimal bioactivity retention in the most polar solvent following organic extraction. Adult assays demonstrated that 50 mg/mL of *M. charantia* will kill adult *A. ceylanicum* within 24 hours, where the same concentration of *L. sativa* will not. These results highlight the potential of *Momordica charantia* as a promising alternative for treating parasitic hookworm infections, addressing the critical need for effective interventions amidst emerging anthelmintic resistance.

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Development of *Eimeria* surrogates to advance control of human *Cyclospora* infections

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Cyclospora causes annual outbreaks of human enteric disease, often associated with consuming contaminated fresh produce. Efforts to reduce public health harms and reputational damage to the fresh produce industry require better understanding of parasite biology and epidemiology, but progress has been slowed by a dearth of parasite material, the inability to propagate the organism in vitro or in any known laboratory animal model, and the danger the organism poses to human health. Here, we outline progress hastened by developing species of *Eimeria* (of poultry) as a biologically sensible and tractable research model, emphasizing recent progress in understanding parasite development, senescence, transcriptional responses to interventions, and prevention by means of simple, cost-effective filters.

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Anticoccidial sensitivity and resistance profiles from commercial turkey flocks in the United States

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Anticoccidial resistance is a growing concern for the US turkey industry due to limited options for control of *Eimeria* in commercial farm settings. From 2019 to 2022, excreta samples were obtained from 24 commercial turkey production flocks to test the sensitivity of *Eimeria* to a panel of six anticoccidial medications including Amprolium (2.5mL/L), Lasalocid (113g/UST), Monensin (90g/UST), Clopidol (225g/UST), Halofuginone (2.72g/UST) and Zoalene (170.3g/UST). After sporulation, young turkeys were inoculated with 1,000 sample oocysts and output was measured for seven day post-inoculation. To determine sensitivity, oocyst output was compared to a non-medicated control group in which a reduction exceeding 79% signified sensitive, 30 - 79% reduction reduced sensitivity, and less and 30% reduction indicated resistance. Sensitivity of samples was the highest for Clopidol with 54.2%, followed by Halofuginone at 33.3%, and all other anticoccidials testing below 10%. Reduced sensitivity of medications ranged from 19.1% - 46.7% of samples. Halofuginone and Clopidol had the lowest incidence of resistance at 20.0 and 20.8%, respectively, followed by Monensin at 47.6% incidence. All other medications had a higher than 50% rate of resistance in field samples. Overall, only 7% of samples were

pan-sensitive, 55% were resistant to three or more anticoccidials, and 14% were pan-resistant. Limited anticoccidial options for turkey producers complicates rotation programs due to reduced downtime before cycling back to a previously applied anticoccidial. Thus, these data highlight the need to adopt different rotation and bioshuttle programs that can limit anticoccidial resistance in commercial settings.

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Phenotypic differences in feeding behavior of multi-anthelmintic drug resistant *Ancylostoma caninum* larvae suggest mutated nicotinic acetylcholine receptor

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Over 500 million people are infected with hookworm, which can result in debilitating and sometimes fatal iron-deficiency anemia. In endemic areas, mass drug administration of anthelmintics must be used frequently due to high rates of reinfection. Although effective, this places significant selective pressure on parasitic nematodes to develop resistance. Resistance to benzimidazoles (BZ), macrocyclic lactones (ML), and pyrantel (THP) in the canine hookworm, *Ancylostoma caninum,* have been documented, including a naturally occurring triple-resistant isolate (BCR) that our group reported earlier this year. We maintain a wild-type isolate (WMD) in addition to the mutant BCR isolate in the lab, which provides an opportunity to compare isolate-specific phenotypic differences. Using an in vitro larval feeding assay, also referred to as an activation assay, we found that BCR infective third stage larvae (L3) resume feeding, or activate, to approximately half the level of WMD L3 in response to host-like conditions. This implies that there is a lesion somewhere in the activation pathway that may be associated with anthelmintic resistance. Pyrantel is a nicotinic agonist anthelmintic that depolarizes the neuromuscular junction of nematodes. Previous experiments in our lab suggest that pyrantel resistance may be related to changes in activation phenotype. When BCR L3 are incubated with standard activation conditions and low levels of pyrantel, activation is increased. To elucidate the relationship between pyrantel resistance and depressed activation phenotype, we tested various compounds that act on muscarinic/nicotinic acetylcholine receptors (mAChRs/nAChRs) to determine the effect on activation behavior in BCR. Our results suggest that mAChRs are involved in larval activation, and while there may not be a nAChR present in the pathway, nicotinic compounds are either interacting directly with the mAChRs or on the muscles involved in feeding. Understanding the mechanisms of anthelmintic resistance is crucial to enhance control tactics, optimize anthelmintic usage, and track the emergence of resistance.

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Demystifying Myxozoans

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The myxozoans are obligate, microscopic parasites known for several economically and ecologically important diseases in fishes. Significant shifts in our understanding of myxozoans have occurred over the past one hundred years, yet there remains much more to learn about these endoparasites. Initially considered protozoans and in their own phylum for many years, these spore-forming parasites are multicellular with conspicuous ‘polar capsules’ (cnidocysts). Taxonomic controversies that lasted decades were resolved with the advent of molecular data in the 1990s, which revealed that myxozoans belong in Phylum Cnidaria, and are thus the diminutive but potent parasitic cousins of corals and jellyfish. With more than 2500 species known, and that number edging inexorably higher, the Myxozoa are likely to number in the tens of thousands, and thus represent the most speciose component of Phylum Cnidaria, out-numbering their more photogenic sisters. Other noteworthy milestones in our understanding of myxozoans include the elucidation of the first life cycle in the 1980s – complex and requiring an alternate annelid host – and which linked previously separate classes of spores. Their vertebrate host range has been discovered to include not only fishes, but reptiles, birds and even terrestrial mammals. Most recently, 'omics has shown myxozoans have some of the most reduced animal genomes, with at least one species having lost its mitochondrial genome completely. Despite genetic simplification, myxozoans exhibit diverse characters that include some species having motile worm-like plasmodial stages and others with motile 'dancing' stages that evade host immune attack. Myxozoan diversification and dispersal have been promoted by human-mediated factors including ecosystem alteration and culturing and translocation of fishes. Climate change and subsequent shifts in vertebrate and invertebrate host ranges are likely to promote emergence of known and novel myxozoan infections and diseases.

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Human Schistosomiasis: From Hieroglyphs to Artificial Intelligence

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Symptoms consistent with human schistosomiasis have been depicted in hieroglyphs on Egyptian tombs, a supposition supported by presence of schistosome eggs in mummies. Napoleon's troops and Napoleon himself were also described with schistosomiasis-like symptoms, including speculation that they contributed to his downfall at Waterloo. Theodore Bilhartz first described human schistosomiasis in 1851 and correctly concluded that pathology is caused by the parasite eggs rather than the worm itself. *Schistosoma haematobium* causes urogenital schistosomiasis and presents as hematuria in less severe infections but can eventually lead to bladder fibrosis, urinary tract blockage, and carcinoma. In women, eggs that become lodged in the cervix and vaginal wall cause female genital schistosomiasis, which is associated with subfertility and increased HIV transmission. Intestinal schistosomiasis is caused by most other species, the two most prominent of which are *S. mansoni* and *S. japonicum.* Intestinal schistosomiasis causes abdominal pain, enlargement of the liver and spleen and may develop into severe liver fibrosis, ascites, and esophageal varices in chronic infections. Schistosomiasis affects an estimated 250 million people in 78 countries, with most cases occurring in Africa. Introduction of praziquantel in the mid-1980s was a critical milestone for schistosomiasis control and has led to impressive reductions in the prevalence of and the morbidity caused by schistosome infections over the last 20 years. While microscopic detection of eggs in the urine for urogenital disease and in stool for intestinal schistosomiasis has long been the mainstay of diagnosis, as infection prevalence and intensity decrease, these methods may miss light infections, leading to underestimated prevalence and missed treatments. Thus, WHO has developed target product profiles for new diagnostic tests for schistosomiasis. Methods under development include detection of parasite antigen or nucleic acid in urine, stool, and blood. While schistosome-specific antibody tests are less useful for monitoring and evaluation of control programs because people remain antibody positive long after successful treatment, recent advances in machine learning to characterize aspects of the antibody response profile beyond specificity have been employed, leading to exciting new possibilities for point of care tests that may contribute to improved schistosomiasis diagnosis, morbidity control, and elimination of transmission.

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A One Health approach to Toxoplasma gondii in the Arctic

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The coccidian *Toxoplasma gondii* is arguably the most ubiquitous parasite, infecting many vertebrate hosts and ranging from the equator to the poles. The ultimate source of *T. gondii* is the intestines of wild and domestic felids, whereas the pathogenicity of *T. gondii* lies primarily in vertebrate intermediate hosts, where it can cross the placenta and affect mammalian fetuses (sometimes triggering abortion), and reactivate in immunocompromised hosts. Unique among coccidians, *T. gondii* can transmit from intermediate host to intermediate host through carnivory. Therefore, while cats are the ultimate source of environmental contamination, the importance of food borne routes of exposure is increasingly recognized. This is almost certainly the case in the Canadian Arctic, where there is high seroexposure to *T. gondii* in wildlife and people, but felids are few and far between above treeline. Over a decade of research in the Canadian Arctic reveals that lynx are a potential source of environmental contamination with *T. gondii*, but are far more commonly infected as intermediate hosts; terrestrial carnivores are good sentinels of circulating levels of *T. gondii,* which are higher in the eastern vs western Canadian Arctic; and exposure to *T. gondii* in the eastern Canadian Arctic is primarily linked to consumption of migratory and aquatic wildlife, rather than terrestrial herbivores like caribou. These findings in wildlife are congruent with human seroepidemiological studies and have significance for Indigenous harvesters, emphasizing the utility of One Health approaches to the complex transmission and ecology of this globally successful, but enigmatic, parasite in the Arctic.

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Are parasitologists obsolete (again)?

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Armand Kuris focused his 2012 Presidential address to the American Society of Parasitologists on the complicated history and geography of human parasites. He emphasized that many of our longstanding parasitic diseases evolved with us in Africa, and most remain there today. As humans migrated out of Africa, they left some of these parasites behind, but acquired a few new ones. Then slave traders moved parasites as well as people, and this allowed several human parasites to colonize the Americas. This was likely the greatest era for human parasites. Early parasitologists stepped up to identify life cycles and test treatments. Subsequent public-health measures, including destroying breeding sites for mosquitos and snails, insecticides, bed nets, screens on windows, toilets, access to medication, and information about hygiene reduced the burden of parasites in wealthy nations. Then with WWII came DDT, and vector borne diseases seemed poised for elimination everywhere. Parasitologists could finally retire. Yet, vectors evolved resistance and bounced back enough that concerns grew and money poured in for vaccine development, with disappointing results. In the 1980s and 1990s parasitologists seemed relevant again, so folks my age got parasitology degrees. We made predictions that global warming and ecosystem disruption should increase infectious diseases further. Instead, most parasitic diseases declined markedly. Such declines indicate that economics has a more powerful effect on human health than climate does. Economic disparity still drives in the global distribution of parasitic diseases, but, assuming global poverty continues to decline, so will human parasites, and probably parasitologists, with fewer courses and careers available. However, war, famine, and displacement can easily reverse this progress, and parasites will be ready to take advantage. Will there be parasitologists there to help?

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Ecology of Fear: ontogeny-mediated non-consumptive effects

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Non-consumptive effects (NCEs) arise in the presence of predators or parasites even when death or infection does not occur and can include changes to host behaviour, physiology, or morphology. A growing body of literature offers evidence of various NCEs in parasite-host interactions, but the focus has been primarily on a single developmental stage of the host. Using the *Drosophila nigrospiracula* – *Macrocheles subbadius* fly-mite system, we investigate the impact of parasite exposure (*sans* infection) during the pupal and adult pre-reproductive stage. First, we exposed fly pupae to ectoparasitic mites—either indirectly (caged mites) or directly (free roaming mites). We found that direct exposure significantly decreased the rate of successful eclosion by 20% (development from pupa to adult) compared to unexposed pupae; however, the duration of pupation was not significantly affected. The indirect exposure did not have a significant effect on either successful eclosion or duration of pupation. Second, we tested how exposing adult female flies to ectoparasitic mites prior to reproduction affected fecundity during the post-exposure, reproductive period. Females indirectly exposed to caged mites had a 35% reduction in the number of offspring produced compared to control flies, but only for the first few days post-exposure; i.e., the effect was reversible after mites were removed. Offspring production returned to levels comparable to the control flies, 4 – 5 days after the removal of mites. The exposure to mites had no significant effect on offspring weight compared to unexposed females. Investigating the diverse NCEs associated with parasite exposure at various life stages of the host is important in understanding the ecology of fear and its total impact on hosts throughout their entire lifespan, with consequences for host ontogeny and population growth.

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Host specificity in hookworms: Species-specific and sex-dependent immune mechanisms mediate host permissiveness

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A long-standing question in parasitology is what mediates the host range of a parasite. Specifically, what is the molecular mechanism that determines why a parasite infects hosts that are permissive to infection, but not those that are non-permissive, and conversely, how does a non-permissive host prevent parasite establishment? Hookworms of the genus *Ancylostoma* demonstrate a wide range of host specificities, ranging from the strict specialist *A. caninum* that can only reliably infect canids, to the generalist *A. ceylanicum* that infects carnivores, humans, and hamsters. Given the broad host range and the ability to infect hamsters, it is surprising that *A. ceylanicum* cannot infect closely related rodents like mice. Evidence suggests that immune mechanisms protect mice from hookworm infection. To test this hypothesis, we leveraged two congeneric species that differ in their host specificity to determine whether the immune system regulates permissiveness to hookworm infection. We found that *A. ceylanicum* completes its life cycle in immunodeficient NSG mice, but not in immunocompetent Swiss Webster mice. However, the closely related host specialist *A. caninum* failed to develop in NSG mice. Neither hookworm species could establish in NOD scid mice, which lack T and B cells yet retain a functional innate immune system, suggesting that the innate immune system restricts infection in mice. Female mice, but not male mice, lacking the signal transducer and activator of transcription 6 (STAT6) supported the development of *A. ceylanicum*, suggesting a sex-dependent mechanism operating in this

mouse strain. Our findings reveal that host specificity in hookworm infections is governed by a complex interplay of immune responses, which are species-specific and exhibit sex-dependent differences.

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Endosymbiotic *Wolbachia pipientis* in Onchocercid Nematodes: The Potential Role of Bacteriophage DNA in the Biology of this Host-Parasite System

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Filarioid nematodes (Nemata: Filarioidea) are responsible for some of the most devastating parasitic diseases of humans and other animals. Within the ovaries of some female filarioids live obligate endosymbionts, α-proteobacteria of the genus *Wolbachia*; these endosymbionts engage in a complex, mutualistic symbiosis with their nematode hosts. Our study investigates the prevalence of bacteriophage elements in *Wolbachia* of filarial nematodes. Previous studies have been unable to identify WO elements in *Wolbachia* of supergroups C, D, J, & L (nematode-specific *Wolbachia*), however, by mapping WO sequences onto available genomes of filarial nematodes, our study finds that WO elements are present in nematode-specific *Wolbachia*. With this new information and the previous findings on the diverse roles played by WO in arthropod *Wolbachia,* we can speculate on the potential impacts of bacteriophage elements within *Wolbachia* of filarial nematodes.

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Stability of the *Heligmosomoides polygyrus* microbiome following host antibiotic exposure

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There is increasing evidence that the parasite microbiome influences parasite transmission and fitness. Characterizing the composition and stability of parasite microbiomes could provide novel strategies for controlling infection. Here we examine the stability of the *Heligmosomoides polygyrus* microbiome following experimental perturbation of the host gut microbiome. We infected C57BL/6 mice (n=24) with *H. polygyrus* and, following parasite maturation, exposed experimental mice to a potent antibiotic cocktail designed to substantially reduce their gut microbiota. For antibiotic-treated and control mice, we collected data on host health, parasite fitness, and microbiota immediately after antibiotic treatment and three weeks post treatment. Antibiotics altered both host physiology and parasite survival. Antibiotic treatment caused a rapid increase in mouse cecum size and a significant reduction in both worm abundance and egg production. To examine how perturbation of the host microbiota altered parasite microbiomes, we are now characterizing parasite and host microbiota using next generation sequencing. These results suggest that optimal fitness in *H. polygyrus* requires an intact host microbiota and will provide insight into the stability of parasite microbiomes.

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An assessment of pathogen risk to juvenile Spring-run Chinook Salmon (*Oncorhynchus tshawytscha*) in the Upper Klamath Basin

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The Upper Klamath Basin (UKB) consists of a diverse assemblage of tributaries, lakes, and reservoirs in Southern Oregon. Historically inhabited by Spring-run Chinook Salmon, hydroelectric dam development in the 20th century cut off all anadromous fish passage to the UKB. With the removal of the four lowermost dams in the Klamath River, Spring-run Chinook Salmon will have access to UKB habitats again. Using sentinel fish exposures, we describe the infection risks that may be encountered upon re-establishment. In spring 2022 & 2023 (once in April and May), replicates of Trinity River juvenile Spring-run Chinook Salmon, raised at Klamath Hatchery, were placed in cages at two sites - a tributary of the Upper Klamath Lake (UKL) and near the outlet of UKL. Within 24 hours post-exposure half the fish in each group were euthanized and immediately necropsied and the other half held in tanks and monitored for disease signs for 28-days, then euthanized and sampled. In 2022 & 2023, *Flavobacterium columnare, Ceratonova shasta*, and *Ichthyophthirius multifiliis* were detected at both UKB sites. *Renibacterium salmoninarum* was only detected in 2022, at both sites. *C. shasta*, *F. columnare*, and *I. multifiliis* were more prevalent in May, associated with higher river temperatures, compared to April. In 2022, fish infected with *F. columnare* and *I. multifiliis* were treated with formalin to prevent mortalities and allow the development of internal parasites. Conversely, in 2023, fish were not treated with formalin to determine the morbidity from *I. multifiliis* or *F. columnare* and *I. multifiliis* co-infections. *F. columnare* and *I. multifiliis* pose a disease risk for out-migrating juveniles in the UKB and throughout the Klamath mainstem, as these two pathogens are expected to be exacerbated with climate change.

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Evaluating the Exposure of Primary School Pupils to Soil Transmitted Ascariasis Through Wastewater and Disposal in Lokoja Kogi State

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The disease of Ascariasis has been around for many years which was first studied by Edward Tyson in the late 17th century. Ascariasis comes in two forms. Children are infected more often than adults. Contamination of surface water with wastewater may occur due to poor infrastructure, such as non-treated effluents, leaky sewage pipes and faulty wastewater treatment plants. The infection is likely to be more serious if nutrition is poor. This study was carried out in Nigeria with particular reference to Lokoja Local Government of Kogi State. Pupils were trained and supervised on how to put the specimen in the poly pots hygienically. Incidence of ascariasis infection among preschool and school aged children was conducted among 100 randomly selected pupils. Pupils' parents/guardian questionnaires respondents on age, sex, family size, source of water supply, nature of liquid /solid/human waste disposal, feeding habits, were 46% farmers and fishermen/women, 18% civil servants and 38% traders. The ova and mature worm loads from the faecal samples collected from the pupils was examined using Ritchie formol-ether concentration technique. The results of incidence of Ascariasis showed % decrease with increasing age, 29.4, 22.4, 16.9, 20 11.8 and 5.3 between ages 2-4, 5-7, 8-9, 10-11, 12 & above respectively. Results showed higher infection rates 56.5% in female and 43.5% in males. The grouping results showed groups with improved water supply, feeding on home prepared meals, use of effective water/liquid/ solid/human waste disposals, had reduced mature worm and ova among pupils. It is recommended that improvement of these conditions will conclusively aid in control of ascariasis.

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Incidence of Intestinal Parasitic Infections in School Children of Faisalabad, Pakistan

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Intestinal parasites are the main reason for morbidity and mortality all over the world particularly in the third world. These parasitic ailments are caused by an environment that is hot and humid, poor sanitation, dirty drinking water, poverty, and illiteracy. Children's poor personal hygiene is considered an important contributing factor to parasitic invasion. In the present study, the prevalence of intestinal parasites and their associated risk factors were determined among the school children of Faisalabad, Pakistan. 150 stool samples were obtained from the assumed children of 3-15 years of age belonging to Private and Government Schools. The various stages of intestinal parasites were identified by using a variety of techniques, including the McMaster technique, the direct smear method, the sedimentation technique, and the formalin ether concentration technique. The prevalence(%) of parasites found were *Ascaris lumbricoides* (4.68), *Giardia lamblia* (4.68), *Entamoeba histolytica* (3.32), *Taenia saginata* (3.90), *Enterobius vermicularis* (3.90), *Trichuris trichura* (2.68) and *Hymenolepis nana* (1.90). *A. lumbricoides* and *G. lamblia* were found to be the most frequently prevalent parasites. Prevalence of intestinal parasites was found to be more among individuals with poor hygiene and uneducated mothers since maternal care plays a vital role in a child's upbringing and maintenance of good health. Educating masses about hygiene and parasitic diseases, along with providing strategies and guidelines to parents for raising children in low socioeconomic circumstances may decrease the load of parasitic infections. Furthermore, school-based deworming is recommended.

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Visualization of environmental cell forms to better understand the ecology and evolution of an important marine pathogen

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Perkinsus marinus is a highly prevalent oyster parasite along the Atlantic coast and has been recognized as a major source of disease and mortality in wild and aquacultured eastern oyster populations for over half a century. While vegetative proliferation of this parasite is well described within the oyster host, the importance of zoosporulation in *P. marinus* epizootiology is far less clear, particularly in light of the life history changes associated with its intensification in the 1980s. To better understand the ecology of this parasite outside of the host, weekly surface and benthic water samples were collected over the year from January 2022 – December 2022, and filtered and preserved on 0.22 membrane filters. Parasite abundance was quantified using qPCR, and cell forms were visualized using fluorescent in situ hybridization. Sampling was repeated in Feb – April 2023 at three sites to investigate potential factors responsible for a recurring spring peak. Environmental *P. marinus* detection closely followed established seasonal infection trends in oyster hosts, with a maximum in late summer/early fall and little detection in winter and spring, with the notable exception of a minor March peak. Visualization of cell forms revealed zoospores and hypnospores to be the predominant stages. The consistent observation of putative zoospores and hypnospores confirms the continued relevance for zoosporulation in *P. marinus* transmission, and a potential significance particularly in supporting the year-round transmission of *P. marinus* that has long been known to occur even during annual periods of low abundance in oyster populations.

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The emergence of *Tetracapsuloides bryosalmonae* in an Oregon salmonid hatchery

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Proliferative Kidney Disease (PKD) is of great economic and ecological concern for cultured salmonids in Europe and North America. It is caused by the myxozoan parasite *Tetracapsuloides bryosalmonae* (*T. bryo*), which alternates between a freshwater bryozoan and salmonid hosts. Although the parasite is native to Oregon, only four occurrences of *T. bryo* infections have been recorded in the state's fish

hatcheries over the past 25 years between 1994 and 2001. That was until 2020, when a PKD outbreak caused severe mortality of juvenile Chinook salmon (*Oncorhynchus tshawytscha*) at Leaburg Hatchery (LBH) on the McKenzie River. In response, and given the parasite's unknown regional epidemiology, we commenced a multi-faceted monitoring program. Monthly from May 2023 to March 2024, water samples from the LBH inflow, raceways and outflow were collected and analyzed for waterborne parasite stages via qPCR, and kidney tissue from both Chinook salmon and steelhead trout (*Oncorhynchus mykiss*) were sampled for histology and PCR. This increased surveillance reveals that the onset of fish exposure to waterborne *T. bryo* spores from bryozoans occurs in early June, and the first kidney infections are detectable mid-July. Disease signs and mortality manifest 6 weeks post exposure when temperatures remain over 15°C, and lead to 100% morbidity as evidenced by swollen kidneys and severe granulomatous inflammation of the kidney in histology. As salmonids are moved among hatcheries and stocked around Oregon, these data can immediately help inform management decisions about when and where to transfer fish to and from this site.

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Developing methods to maximize lab infection success of *Echinostoma trivolvis* lineage C in gastropod first intermediate hosts

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Completing parts of trematode life cycles in the laboratory is a useful way to obtain experimentally infected hosts and identify how specific aspects of parasitism influence host ecology and behaviour. However, these efforts can be hampered by unknown host specificity and the uncharacterized influence of host and parasite factors on infection success. *Echinostoma trivolvis* lineage C is a genetically distinct trematode within the *E. trivolvis* species complex that has only been identified by DNA sequences from adult and larval stages recovered from naturally infected muskrats (*Ondatra zibethicus*) and the marsh pondsnail (*Ladislavella elodes*), respectively. To promote its use in laboratory research, we determined the effect of host species/morphotype, host size, and miracidial dose on the infection success in potential first intermediate hosts. In laboratory, we exposed 2 freshwater snail species (*L. elodes* and *Planorbella duryi*) as well as a morphological variant of *L. elodes* (formerly known as *Stagnicola reflexa*) to 2 miracidia to determine specificity in the first intermediate host. Among these 3 snail groups, we also evaluated the effect of host size on infection success using 3 size classes (1 – 5 mm, 5 – 10 mm and 10 – 15 mm) and a dose of 2 miracidia. Within *L. elodes*, we compared the effect of 2 doses (2 and 5 miracidia) on infection success. Rediae and cercariae developed within one host species, *L. elodes,* as well as its *S. reflexa* morphotype. However, prevalence in *L. elodes* was higher than *S. reflexa,* with the highest prevalence in small and medium sizes of *L. elodes.* At the higher dose of miracidia, prevalence increased in small and medium-sized *L. elodes* relative to the low dose. Our results confirm the first intermediate host species observed in nature but indicate that prevalence is influenced by host species morphotype, host size and parasite dose. To maximize the number of infected snails in laboratory, we recommend exposing medium-sized *L. elodes* to 5 miracidia, though small *L. elodes* are also suitable. This research encourages further use of *E. trivolvis* lineage C in the laboratory to explore aspects of host-parasite interactions such as parasite-modified behaviour.

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Trichinella control: a singular public health achievement

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Before the advent of effective protective measures, most cases of human trichinellosis derived from consumption of undercooked, infected pork. Research conducted at the United States Department of Agriculture (USDA) since 1891 has helped to reduce *Trichinella* infections in conventionally-raised domestic pigs to negligible levels, as established by recent conclusion of an ambitious national survey of animals raised under biosecurity safeguards defined by the Pork Quality Safety + production standard. Here, we review the history of this scientific progress, placing special emphasis on work conducted at the USDA and paying tribute to Dr. K. Darwin Murrell for his uniquely consequential success as a scientist and science administrator, conducting and inspiring research that resulted in this singular public health success.

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One year follow up of a cross-sectional study of hookworm infection and response to deworming in Beposo, Ghana.

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Hookworms are soil-transmitted helminths that infect over 500 million people worldwide, primarily those in low-income countries. Infection with hookworm can cause anemia, malnutrition, and stunted physical and cognitive development. Deworming campaigns have attempted to combat hookworm infections in endemic areas for decades. However, there is growing variability in the effectiveness of deworming drugs, and post-treatment reinfections are abundant. There are significant gaps in our understanding of critical host-parasite factors that mediate hookworm infection, reinfection, and response to deworming treatment in endemic communities. A five-year prospective study was undertaken to better understand drivers of hookworm infections in rural Ghana and thus improve future intervention strategies. Here, we present the year 2 cross-sectional assessment of the human behavioral, demographic, and social factors influencing community infection levels and response to treatment in Beposo, Ghana. Among 588 participants >3 years old recruited in year 1 (2022) of the community-wide survey, we collected demographic data, social behaviors, and sanitation infrastructure. In June-July 2023 we collected anthropometric measurements and outcomes of interest, including hookworm infection status, eggs per gram of stool (EPG), and treatment success following administration of 400 mg of albendazole, including cure rate (CR) and egg reduction rate (ERR). Univariate analysis was performed to describe participant characteristics and potential risk factors. Multivariable logistic regression was used to assess the relationship between exposures, infection risk, and treatment outcomes and was adjusted for potential confounders. The prevalence of hookworm was 31.9%, and the CR was 58.5%. The baseline mean EPG was 343.6 (+/- 405.9), and the post-treatment EPG was 49.6 (+/- 98.2). The overall ERR was 80.40%. Analysis revealed that age, BMI, gender, owning agricultural land, and lack of electricity were factors significantly associated with increased odds of hookworm infection. Using boreholes as water sources was significantly associated with increased odds of hookworm infection and higher EPG before treatment. Subjects with a higher pre-treatment EPG were significantly less likely to be cured following albendazole administration and had a lower ERR. These findings suggest that 400 mg of albendazole may be insufficient to treat heavy parasite burdens.

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Insights on the transcriptome analysis of the *Taenia solium* development from metacestode to juvenile - adult

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Taenia solium is a parasitic flatworm responsible for taeniasis and cysticercosis. Neurocysticercosis, caused by ingesting eggs shed in human feces, is a leading cause of acquired epilepsy worldwide, particularly in areas with poor sanitation. The infection process begins when humans eat pork contaminated with the parasite's larval forms (cysticerci). Bile salts and digestive enzymes trigger the parasite's scolex to emerge from its vesicle. Generally, only one larva attaches to the epithelium of the small intestine and grows into an adult tapeworm through a process called strobilation. This process involves the continuous generation and maturation of new body segments from behind the scolex. Definition of the anterior/posterior (AP) axis orients evagination of the scolex and strobilation. The mature segments release eggs into the host's feces, continuing the cycle of infection. Despite advances in cestodes like *Echinococcus*, *Mesocestoides*, and *Hymenolepis*, the molecular pathways governing *T. solium*'s development remain incompletely understood, including the transition from larval stages into the juvenile and adult forms. We employed Illumina paired-end RNA sequencing to examine changes in gene expression related to key developmental pathways Notch, Wnt, and Hedgehog in pools of cysts grown in the presence and absence of taurocholic acid (TA), a bile component. Cysts were categorized according to morphology, for comparison: non-evaginated (6 h after culture), early evaginated (1-2 days in culture), and late evaginated (5 days in culture); we also sequenced cysts before culture. The AP axis was being established in cysts before culture, which showed the highest levels of anterior (sfrp) and posterior (wnt1) marker genes among all groups, regardless of TA presence. The role of TA in accelerating evagination is evident by the higher expression of wnt1 in early as opposed to late evaginated cysts. The expression of wnt11a, which plays a critical role in strobilation at the posterior end, was significantly higher in late evaginated cysts than in all other groups. Antagonic components of the Hedgehog pathway were also differentially expressed: ci was higher in cysts before culture, and slimb in early and late evaginated cysts. Our findings confirm that posteriorization drives *T. solium*´s development, similar to other tapeworms.

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Malaria vector control with indoor residual spraying in students' hostels at Awka - Nigeria

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Human malaria, caused by *Plasmodium* species and transmitted through bites of infected female *Anopheles* mosquitoes, remains life-threatening in Nigeria. It is a major reason for undergraduates' absenteeism from lectures. One of the strategies employed for malaria control in the country is indoor residual spraying (IRS) with insecticides capable of knocking down in-door resting mosquitoes to disrupt malaria transmission cycle. A cross-sectional study in undergraduate hostels at Awka was undertaken to identify malaria vector species, as well as estimate their indoor resting density (IRD) and man biting rate (MBR). Pyrethrum knockdown (PKD) method was used to sample indoor resting mosquitoes between 6am and 9am in 16 rooms where 28 undergraduates slept during their first 5 nights on resumption for 2023/2024 academic session. Knocked-down mosquitoes were morphologically identified based on appearance of their head, thorax, wing colors, and tarsal segments of hind legs; and subsequently categorized by close observation of their abdomens, as blood-fed or not. Data generated were subjected to descriptive statistic while IRD and MBR were estimated using their respectively derived formulae: IRD = [(Total number of mosquitoes collected / number of rooms sampled) / Total number of nights] , and

MBR = [(Total number of engorged mosquitoes collected / number of rooms' occupants) / Total number of nights]. All the 128 mosquitoes knocked-down were identified as *Anopheles* species but 93 (72.7%) of them were engorged while the abdomens of 35 (27.3%) contained no blood meal. Estimated IRD was 1.6 mosquitoes per room per night while MBR was 0.66 bites per man per night. High percentage of engorged mosquitoes observed indicated intense activities of malaria vectors in hostels. Result also demonstrated that IRS with pyrethrum-based insecticide was still effective in knocking down mosquitoes. However, because of potential danger of insecticide-resistance development, as well as exposure to non-target organisms, it is recommended that future research should be focused on development of newer insecticides, improvement in application techniques, and implementation of feasible monitoring and evaluation strategies for malaria vector management in the area.

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Shifts in trematode parasite community structure from headwaters to a mainstem river in the Willamette River Basin, Oregon

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The factors shaping parasite community structure have long been a topic of interest. Dispersal limitation, abiotic factors, and species interactions have been posited as important drivers of parasite diversity and composition, but relatively few studies have tested these drivers concurrently. We sampled trematode parasites from freshwater snails (*Juga plicifera*) at 137 stream sites within the Willamette River Basin in western Oregon, USA. The sites spanned a gradient of stream order, from 1st order headwaters to the 8th order mainstem Willamette. We predicted that trematode richness and composition would shift over the stream gradient in relation to environmental factors, stream network connectivity, and potentially due to interactions between co-occurring trematodes. Using a combination of morphological methods and 28S ribosomal DNA sequencing, we identified at least ten taxa of trematodes infecting *Juga* snails in the watershed, which have been preserved at the Museum of Southwestern Biology's Division of Parasites. Richness peaked in mid-order stream reaches and was lower in headwaters and the mainstem river. Trematode composition shifted in relation to stream order, with distinct communities in headwaters versus the mainstem. These shifts were correlated with environmental factors which may have direct effects on trematode transmission, or they may coincide with shifts in distributions of secondary- or definitive hosts which are needed to complete trematode life cycles. We detected a role of trematode-trematode interactions in shaping site-level infection patterns, but these interactions appear to be less important in driving community structure at the watershed scale. Lastly, we found some support for the idea that more isolated sites had more unique communities, suggesting a role of dispersal limitation. Taken together, our results indicate that trematode community structure in river ecosystems is shaped by a combination of ecological factors, which likely operate over unique spatial scales.

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Seasonal succession of cestode metacommunities in two North American *Sorex* shrews

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Patterns in parasite diversity and prevalence are determined by a suite of factors ranging from environmental conditions like temperature and precipitation to host behavior. The forces shaping these patterns are increasingly unstable due to anthropogenic climate change, emphasizing the importance of leveraging natural history collections to understand historical contributions of environmental conditions to the spatiotemporal distributions of parasitic diseases. We implemented frameworks from metacommunity analyses to test for seasonal trends in parasite community structure attributable to host environment in two North American shrew species, *Sorex cinereus* and *Sorex monticola*. Shrews were collected using snap traps and pitfall traps periodically from 2009 to 2018 at two sites in Cowles, New Mexico. Whole GI tracts were removed from the shrews, and tapeworm taxa were identified to genus using high-throughput amplicon sequencing of *28S rRNA* gene. Evaluation of metacommunity structure (EMS) revealed coherent, dynamic cestode metacommunities indicative of individual parasite responses to seasonal trends in climate variables. We used boosted regression trees to identify latent variables predictive of cestode metacommunity structure and identified "day of the year" as having the greatest relative influence of parasite community structure, followed by precipitation and host body size. This work demonstrates for the first time the utility of the EMS framework for detecting fine-scale seasonal dynamics in parasitic helminth communities.

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Greater than the sum of its parts: integrating DNA metabarcoding and classical methods in parasitology to better understand parasite community dynamics

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With ongoing anthropogenic changes to the climate and environment, there is a growing need to incorporate infectious diseases into assessments of wildlife and ecosystem health. Necropsies, fecal analyses, visual encounter surveys, among others, are classical approaches used to survey the community structures and dynamics of parasites and pathogens. While these practices are reliable and broadly implemented, they can be invasive, potentially lethal, and often necessitate specialized training. Recent advancements in high-throughput sequencing, such as DNA metabarcoding, can ameliorate some of these challenges. All organisms shed DNA fragments into their environment via feces, mucus, skin, and other pathways; we can take advantage of this DNA to indirectly measure target organisms without capture and sacrifice. In many studies of free-living organisms, DNA metabarcoding has enabled faster, non-invasive, and complementary or more comprehensive results than classical methods. While DNA metabarcoding has been increasingly applied to ecological studies, and more recently to parasitology, this method is also not without weaknesses. Current limitations include a lack of sequenced taxa deposited in online databases that constrains detectability, the difficulty of developing effective primers that captures the range of taxa inherent to metabarcoding studies, as well as the inability to accurately quantify biomass based on DNA reads. For ecological parasitology, we can integrate classical parasitology approaches with DNA metabarcoding to achieve results with higher resolution across larger scales compared to using a single method in isolation. Already DNA metabarcoding has been combined with fecal analysis to monitor the helminths infecting the endangered Southern Resident Killer Whale (*Orcinus orca*) population of the pacific northwest. Similarly, fecal analysis and necropsies were combined with DNA metabarcoding to expand host records and capture the helminth community of the San Miguel Island fox (*Urocyon littoralis littoralis*), a species

vulnerable to extinction. The transformative impact of DNA metabarcoding in ecological research highlights its vast potential. By taking an integrated approach that combines both traditional and innovative methods, we can deepen our understanding of parasite dynamics and their implications for host and ecosystem health.

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Characterizing bias, error, and quantitative potential in 18S metabarcoding of parasite assemblages

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Recent advances in DNA sequencing technology, particularly metagenomic barcoding (metabarcoding) have made high-throughput collection of parasite data over time and space feasible. Although this type of sequencing has been thoroughly studied and standardized where applied to other organisms, such as bacteria, metabarcoding parasites is relatively new and unstandardized. Furthermore, evidence from other eukaryotic metabarcoding systems demonstrates that error and bias are common in pipelines and can complicate data interpretation, leading to inconsistent conclusions. To address these issues, we used three complimentary approaches. Using engineered mock community standards with different organism distributions, we measured and compared levels of PCR bias associated with commonly published 18S V4 primer sets. We also systematically identified sources of error and inconsistency in our 18S metabarcoding pipeline steps. Finally, we assessed the quantitative potential of our assay by comparing 18S sequencing read numbers to fecal egg counts for common soil-transmitted helminths of human medical importance. We found that 18S V4 primers have a wide range of PCR bias with VESPA primers having the lowest in both mock communities. We also identified potential for error across several steps of the pipeline, particularly DNA extraction and data analysis, and found that the choice of DNA polymerase and PCR conditions have dramatic effects on end results. We therefore make recommendations for a standard protocol to minimize bias and describe the use of community standards to monitor metabarcoding quality. Finally, we demonstrate promising potential for soil transmitted helminth quantification using VESPA metabarcoding, although further work is needed to characterize the relationship between read number and biomass in fecal samples.

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Host Traits, Abiotic Variation, and Community Similarity: Parasite β-Diversity Across Life Stages and Scales

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Drivers of community similarity affect assemblage-level changes that lead to differences in species diversity between habitats; thus, they provide key insight into biodiversity patterns across multiple scales. Concepts of β-diversity originally developed for use in free-living communities have been widely applied to parasite communities to gain insight into how infection risk changes in different ecological settings by comparing parasite communities across abiotic and biotic gradients. Yet, key knowledge gaps, such as links between β-diversity of parasite communities at different life stages, remain. Gastrointestinal nematodes hosted by African buffalo provide a useful example for comparing drivers of adult and larval community similarity. Within-host traits significantly affect adult nematode β-diversity, while extra-host variables significant affect larval β-diversity. Meanwhile, examination of adult and larval communities from the same host provides evidence that within-host traits also affect the larval

community indirectly via the adult community; thus, linking drivers of β-diversity between these two life stages. This example provides a framework for comparing community-level processes acting on adult parasites with those acting on their free-living immature stages. This, in turn, provides context for infection risk and the persistence of parasite assemblages, both of which are useful for predicting and preventing parasitic diseases.

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Molecular characterization of a *Sarcocystis bovifelis* -like sarcocysts in American beef

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Infections by the species of the apicomplexan genus, *Sarcocystis* are ubiquitous in cattle worldwide. There is considerable debate concerning the identity of *Sarcocystis* spp. in cattle. Proper diagnosis of *Sarcocystis* spp. is important to assess their economic and public health importance. Currently there are 8 named species: *Sarcocystis hirsuta, S. cruzi, S. hominis, S. bovifelis, S. heydorni, S. bovini, S. sigmoideus*, and *S. rommeli*. Of these, sarcocysts of 2 species, *S. cruzi*, and *S. heydorni* have thin (< 1µm) cyst walls whereas sarcocysts of other bovine species have thick (>3µm) cyst walls. Additionally, there are unnamed *Sarcocystis* spp. Two species, *S. hominis* and *S. heydorni,* are zoonotic. Of these bovine species of *Sarcocystis,* the full life cycle is known only for *S. cruzi*. *Sarcocystis cruzi* (transmitted via canids), the most pathogenic species which causes abortion, low milk yield, poor body growth, and outbreaks of clinical sarcocystosis and death. The thick walled sarcocysts have been often associated with an inflammatory condition of striated muscles (termed bovine eosinophilic myositis; BEM), leading to condemnation of carcasses at slaughter. One difficulty with definitive molecular identification of bovine *Sarcocystis* species is that there is no DNA available from experimentally infected cattle. Here, for the first time, we characterized a thick walled *Sarcocystis* species from American beef using molecular tools. During a survey of American beef for *Sarcocystis* infections, thick (cyst walls ~ 5µm) walled sarcocysts were found in 1 of the 200 beef samples examined from grocery stores. Its sarcocysts were microscopic (< 1 mm long). Molecular characterization employing several genetic markers (18S rRNA, 28S rRNA, COX-1, ITS-1, GAPDH-1, RON-1 and RON3) revealed 100% identity with *Sarcocystis bovifelis*-like sarcocysts from Europe. In summary, as yet there is no evidence of zoonotic *Sarocystis* species in American beef.

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High, but variable prevalence of *Sarcocystis* infections in the American bison (*Bison bison*) beef destined for human consumption

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According to the National Agricultural Statistics Service Census of Agriculture of 2022, bison meat production and consumption has been increasing within the past 5 years in the US. Bison (*Bison bison*) and cattle (*Bos taurus*) are closely related (can interbreed) and they share many parasitic fauna, including those in the apicomplexan genus, *Sarcocystis*. Cattle are commonly infected with one or more of the 8 named species: *Sarcocystis hirsuta, S. cruzi, S. hominis, S. bovifelis, S. heydorni, S. bovini*, *S. sigmoideus,* and *S. rommeli*. Two species*, S. hominis* and *S. heydorni*, are zoonotic. Of these, *S. cruzi* and *S. heydorni* form thin walled (< 1µm thick) sarcocysts whereas remaining species have thick-walled (> 3µm thick) sarcocysts. Among these, the full life cycle is known for only *S. cruzi* (transmitted via canids); it is recognized as the most pathogenic species, causing abortion, low milk yield, poor body growth, outbreaks of clinical sarcocystosis and death and it has been transmitted experimentally to bison. Unlike in cattle, nothing is known of the prevalence of *Sarcocystis* infections in bison raised for human

consumption. We therefore tested 197 bison from 3 sources (Nebraska 141; South Dakota 36; New Jersey and Pennsylvania 20). Frozen tongues were purchased and examined histologically. Thin-walled sarcocysts, presumed to be *S. cruzi*, were detected in 121 of 141 (85.8%) tongues from Nebraska, 35 of 36 (97.2%) tongues from South Dakota. Strikingly, none of the 20 tongues from New Jersey or Pennsylvania were infected. The bison in Nebraska and South Dakota were raised on open range and were grass fed, whereas bison in New Jersey were raised in semi open range and their diet was supplemented with grain and high-quality hay, perhaps accounting for the differences in prevalence of *Sarcocystis*. Molecular and ultrastructural studies are in progress to determine species of *Sarcocystis* in bison.

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Surveillance of the Zoonotic Parasite *Echinococcus multilocularis* in New York State Suggests Multiple Introductions of the European Variant in Wild Canid Populations

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Echinococcus is a genus of cestode parasites of veterinary and medical importance globally. Two species, *Echinococcus granulosus s.l.* and *Echinococcus multilocularis*, are endemic to North America and are the etiologic agents of cystic echinococcosis and alveolar echinococcosis, respectively. We first detected *E. multilocularis* in New York in 2013 through opportunistic genetic testing of scat samples from a wildlife habitat study. Since then, we have initiated surveillance studies to gather data on the occurrence of this parasite in New York. Our surveillance for the parasite in the canid definitive hosts from 2019-2023 yielded 8 detections of the parasite. All of these, and the earlier cases, were determined to represent the European variant of *E. multilocularis* based on DNA sequence analysis. The European (EU) variant is believed to have greater pathogenic potential and transmissibility than the endemic North American (NA) variant, increasing the concern for spillover from wildlife to humans. To facilitate screening of clinical samples, we also developed a rapid SNP PCR test, based on a simple conventional PCR method, to differentiate the EU and NA variants without the need for DNA sequencing confirmation. Additionally, all of our positive samples were genotyped at the EmsB microsatellite region of *E. multilocularis*, revealing several EmsB genotypes that are functionally identical EmsB genotypes from EU variants in France and Switzerland. The New York specimens were distinct from profiles of the EU variants found Western North America. These data suggest that multiple genotypes of *E. multilocularis* occur in New York, and that these represent one or more unique introductions from Europe rather than a spread from western regions of Canada and the United States where other EU variants are found. Our results point to the complex epidemiology of the parasite in North America and warrant further detailed surveillance to evaluate the risk for human acquisition with this zoonotic parasite.

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Species diversity and host specificity at the wildlife-farm animal interface

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Grazing animals are exposed to a multitude of gastrointestinal nematode (GIN) parasites. Although it is known that different parasites have different host specificity, the horizontal transmission between different hosts is not well investigated using molecular tools. The horizontal transfer can spread parasite strains that are resistant to anthelmintic drugs that are commonly used as a prophylactic to keep the parasite burden low on pastures. In this study we have sampled roe deer, fallow deer, red deer, reindeer, mouflon, and sheep across Sweden. Parasite eggs in the feces were counted and. after

extracting parasite DNA from larval cultures, metabarcoding of the ITS2 region was performed using PacBio sequencing technology. Nematode eggs were found in ⅔ of roe deer and reindeer, ½ of fallow deer, ⅛ of red deer and in all sheep. From the metabarcoding, the greatest GIN species diversity in wildlife was found in roe deer (n = 24), followed by fallow deer (n = 19) and red deer (n = 20), mouflon (n = 10) while only eight GIN species were found in reindeer. In domestic sheep 16 species were found, of which *Haemonchus contortus* was the most abundant species and it was also found in some of the wildlife species. Across all host species, the most common generalist found was *Trichostrongylus axei*. The findings indicate that wild animals in Sweden are infected with species that could be transmitted to sheep. However, the risk of this to happen is low as the GIN species profiles between host species are so different.

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How diagnostic are ribosomal ITS markers for schistosome species identification and characterization of putative hybrids?

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Diagnostic markers for species identification require validation against panels of morphologically or genomically characterized samples from across the species range before they can be used with confidence. We provide a cautionary tale about the dangers of using putatively diagnostic markers without prior validation. SNPs within ribosomal ITS have been extensively used to differentiate between *Schistosoma haematobium*, a fluke parasite of humans, *S. bovis*, and *S. curassoni*, related parasites infecting cattle or goats. Furthermore, parasite larvae showing double peaks at putatively diagnostic SNPs from Sanger sequencing chromatograms have often been interpreted as evidence for first generation or recent hybrids. We Sanger sequenced both ITS1 and ITS2 from 286 parasites (209 from humans, 28 from cows, and 49 from snails) collected from 13 African countries. Additionally, we sequenced whole genomes (208) or exomes (78) for these samples to compare the ITS genotype with whole genome ancestry for the same sample set. We used 24.6M commonly genotyped SNVs from the whole genome data to define two well differentiated species (FST > 0.7), with 269,779 SNPs showing perfect concordance with species identify in our sample set (*S. haematobium* and *S. bovis*). The ITS markers performed well for determining species identity in 11/13 countries. However, the ITS markers show incomplete fixation in some West African countries. Notably, 24/64 (37.5%) genomically defined *S. haematobium* from Nigeria, and 5/111 (4.5%) from Niger carry four putatively diagnostic ITS2 SNPs (ATAT at bases 704, 759, 809 and 879) from the *S. curassoni* reference that are commonly used in genomic epidemiology surveys. 14 parasites carried mixed peaks at putatively diagnostic ITS SNPs suggesting recent hybrids; however, the genomic data clearly indicate that these parasites are *S. haematobium.* These results show that while ribosomal ITS markers perform well in many African countries, they are unreliable markers for differentiating species or identifying recent hybrids in West Africa. We caution against reliance on single locus markers and conclude that genome sequencing or abbreviated genotyping of a subset of informative SNPs can provide a more robust approach to differentiate species and evaluate evidence for hybridization in schistosomes.

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Using Population Genomics as a Tool to Evaluate Global Helminth Control Programs

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Mass Drug Administrations (MDA) of anthelmintic drugs to control helminth infections represent the largest drug distributions programs in the world. These global MDA campaigns reach >700 million people annually and distribute billions of doses of drugs each year. Though these drugs act directly on worms, little is known about their impact on helminth population dynamics. Instead, current metrics for program efficacy revolve solely around the number of doses distributed and the clinical measures from treated individuals. Population genomics using whole-genome sequence data is a powerful approach to track worm populations following exposure to anthelmintic drugs. If these drugs remain effective and MDA coverage is sufficient, repeated exposure through MDA should act as a strong selective pressure. Determining how these drugs affect helminth populations across time and space through a population genomic and genomic epidemiological framework therefore represents a novel and important tool to evaluate program efficacy. Here, we explore the untapped potential of applying an "eradication genomics" framework to the problem of plateauing treatment effectiveness, specifically for albendazole and *Necator americanus* infection in Ghana. We will discuss the crucial role helminth population genomics can play in global health campaigns and the challenge of implementing these strategies in real-world settings.

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Mitogenome organization, diversity, and evolutionary relationships of proteocephalidean tapeworms (Cestoda, Onchoproteocephalidea) unveiled by a genome skimming approach

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Proteocephalidean tapeworms (Onchoproteocephalidea I) are a diverse group of acetabulate cestodes that primarily infect bony fishes, snakes, and lizards worldwide. While molecular data, mainly from Sanger sequencing of the nuclear 28S rRNA and the mitochondrial MT-CO1, has been crucial to a better understanding of the taxonomy and systematics of proteocephalids, long-standing questions on the evolution of this taxon remain to be answered. Here, we subjected ethanol-preserved proteocephalid specimens to high-throughput DNA sequencing, using a genome skimming approach, to unravel the mitogenome organization and diversity of dozens of species. Posterior most proglottids of vouchered specimens were provided by the Natural History Museum (Platyhelminthes collection), Geneva, Switzerland (MHNG-PLAT), and the Institute of Parasitology, Biology Centre of the Czech Academy of Sciences, České Budějovice, Czech Republic (IPCAS). Of 96 Illumina sequenced libraries, 90 yielded

complete or partial mitogenomes. These 90 mitogenomes correspond to 83 species in 40 genera from all continents except Antarctica, including parasites of fishes (87%), snakes, monitor lizards (Varanidae), and the common opossum *Didelphis* marsupialis (the only mammal known to host proteocephalids). The resulting mitogenomes varied from 13,522 to 14,008 bp, encompassing 12 protein-coding genes, 22 transfer RNA genes, 2 ribosomal RNA genes, and 2 non-coding regions, with identical gene arrangements. The newly characterized mitogenomes will be used to assess the evolutionary relationships of proteocephalids under a total evidence framework, and the outcomes will be presented at this meeting.

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New data, new insights: A revised phylogeny of the Lecanicephalidea

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The most recent phylogeny of the elasmobranch tapeworm order Lecanicephalidea was based on sequence data from four loci for 61 species in 22 of 25 valid genera and three undescribed genera. The resulting topology supported the eight families Aberrapecidae, Cephalobothriidae, Eniochobothiidae, Lecanicephalidae, Paraberrapecidae, Polypocephalidae, Tetragonocephalidae, and Zanobatocestidae. Subsequently, unpublished data for the 28S rDNA gene for two of the remaining three genera (i.e., *Healyum* and *Quadcuspibothrium*, both from devilrays) suggested these genera represent a ninth family. Over the past five years, we capitalized on several decades of global collections of cestodes from batoids and have generated a 402-loci data set using next generation sequencing for nearly 350 lecanicephalideans representing all but one of the 26 currently valid genera. A preliminary phylogenetic tree confirmed *Healyum* and *Quadcuspibothrium* as a distinct family. It also provided evidence for the existence of at least two additional new families. One is a clade comprising specimens provisionally referred to as “New genus 13” from eagle rays in the genus *Aetomylaeus* and specimens representing yet another new genus from stingrays in the genera *Neotrygon* and *Taeniura*. Furthermore, *Tylocephalum* was not recovered within the Cephalobothriidae, suggesting it should be assigned to a distinct family. The preliminary phylogenetic tree also supports taxonomic actions below the family-level. We confirmed *Adelobothrium* and *Cephalobothrium*, both exclusively parasitizing Pacific eagle rays, to lack reciprocal monophyly and thus requiring revisionary work. *Staurobothrium*, erected in 1905 and essentially considered a *genus inquirendum* since, actually appears to be valid. Despite all of its members bearing an unusual lateral expansion of proglottids on their immature strobila, the genus *Hornellobothrium* appears to be non-monophyletic. Most problematic is the fact that specimens identified as belonging to the genus *Polypocephalus* because of the presence of tentacles on the apex of the scolex were confirmed to be wildly polyphyletic if the morphologically cohesive and distinct genera *Anteropora*, *Seussapex*, and *Corollapex* are to be retained. As a result of the more than seven-fold increase in taxon sampling in the polypocephalids, a number of clades of “*Polypocephalus*” have emerged, greatly facilitating the search for diagnostic features for each phylogenetically supported subgroup.

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Machine learning models accurately predict clades of proteocephalidean tapeworms (Cestoda, Onchoproteocephalidea) based on host and biogeographical data

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We reviewed the phylogenetic relationships of tapeworms with focus on the proteocephalids (Onchoproteocephalidea I) using publicly available (671) and newly generated (90) sequences of 28S rRNA and the mitochondrial MT-CO1 for 537 terminals. Tree search was conducted in TNT v1.6 under the parsimony optimality criterion using a total evidence approach. Interestingly, we were not able to recover the reciprocal monophyly of Proteocephalidae and one of the taxa used as outgroup, *Acanthobothrium*. In addition, it was difficult to reconcile the tree with individual data representing biological, ecological, and zoogeographical traits of the hosts and parasites using traditional character optimization strategies. To test the predictive potential of combined (not individual) host and biogeographical data in the context of the proteocephalid tree, we trained Random Forest machine learning models and demonstrated that they are capable of correctly placing 89% of the terminals into eight representative clades. Furthermore, we interactively perturbed the tree at increasing levels of perturbation probability and observed that model accuracy correlates negatively with the degree of clade perturbation. Our analyses show that even the host and biogeographical data, which individually correlate poorly with the tree, can be used to accurately predict proteocephalid clades when analyzed together. This is to be expected if the evolution of proteocephalidean clades depends on host and biogeographical attributes. We discuss how these machine learning models may serve to provide external support for unpuzzling the proteocephalid tree.

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Morphological and Molecular analysis of *Mathevotaenia* (Cestoda: Anoplocephalidae) from the Giant Anteater, *Myrmecophaga tridactyla* (Pilosa: Myrmecophagidae) In Colombia

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We present a morphological and molecular analysis of tapeworms in the genus *Mathevotania* (Cestoda: Anoplocephalidae) obtained from a Giant Anteater, *Myrmecophaga tridactyla* (Pilosa: Myrmecophagidae), collected in Casanare Department, Colombia. A phylogeny generated from morphological data of the South American species of *Mathevotaenia* is presented. The host was injured by civil personnel in the wild and received for medical assistance at La Palmita Research Station (Trinidad, Colombia) during a field parasitology course in 2019. Due to a poor prognosis, the animal was euthanized. Adult tapeworms were obtained from the small intestine, then processed and stored for morphological and molecular analysis. *M. tetragonocephala* is the only species of anoplocephalid cestode recorded from anteaters. The morphology of the analyzed specimens differs from all congeners infecting mammals in the Americas by the number of testes per segment, cirrus pouch size, genital atrium dimensions, and the disposition of its complex osmoregulatory system. This is the first report of *Mathevotaenia* genus infecting the Giant Anteater in Colombia, and the first genome sequence obtained from the genus in the Americas.

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Progress in the phylogeny and classification of the Rhinebothriidea

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The elasmobranch cestode order Rhinebothriidea was erected in 2009 and contains species that are primarily parasitic in batoids (i.e., stingrays, skates, and guitarfish). An ongoing targeted gene capture global survey for 402 protein-coding loci is in progress on cestode samples from across the rhinebothriidean phylogenetic landscape. Comprehensive species assemblages from all six rhinebothriidean families are included in the study: Anindobothriidae, Anthocephaliidae, Echeneibothriidae, Escherbothriidae, Mixobothriidae, and the Rhinebothriidae. An estimated total of 185 rhinebothriidean species have been included in the current analyses, and 68% of these species are thought to be new to science. Species of 14 of the 23 known rhinebothriidean genera are included. The sequence data support the monophyly of the order as a whole, as well as each of these families. In addition, the analysis provided evidence for two clades that represent genera new to science, and a clade for which the genus *Pillersium*—considered *incertae sedis* for nearly a century—is appropriate. Genera such as *Barbeaucestus*, *Pseudanthobothrium*, and *Rhinebothroides* will need to be synonymized to preserve monophyly of *Anthocephalum*, *Echeneibothrium*, and *Rhinebothrium*, respectively. In addition, there is considerable phylogenetic diversity in genera such as *Anthocephalum*, *Rhinebothrium*, and *Stillabothrium* and, in the future, new genera could be erected to formally recognize monophyletic subgroups with distinct morphologies. Patterns of host use and biogeography are emerging from the data. For example, within *Rhinebothrium sensu stricto,* the species infecting freshwater hosts do not form a monophyletic group. Instead, there are two freshwater clades: One with species infecting freshwater stingrays in South America (including species of *Rhinebothroides* and *Rhinebothrium*) and another with species infecting freshwater stingrays in Southeast Asia, indicating two independent events of colonization of freshwater hosts within the same genus. Phylogenetic hypotheses that are emerging from the targeted gene capture survey will allow us to devise a robust, phylogenetically-informed classification system for the Rhinebothriidea.

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How many species of avian haemosporidian are there?

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Over the last 25 years, there has been an explosive increase in research on the haemosporidian parasites that infect birds. Many hundreds of studies focused on avian haemosporidians have been published in this time period, addressing fundamental questions in parasitology and the ecology and evolution of infectious diseases. Early molecular studies of avian haemosporidian diversity estimated that the number of avian haemosporidian species may approach the estimated number of extant bird species (~10,000), yet the question of avian haemosporidian species richness has not been formally addressed. Here, I attempt to answer a series of questions regarding the global richness of avian haemosporidian parasites. First, I ask whether even the most widely studied and thoroughly sampled bird species on earth have been completely inventoried for the haemosporidian species that infect them. Next, I ask whether we have come close to characterizing the haemosporidian community richness of any single region on earth. Lastly, I attempt to estimate the richness of avian haemosporidians across North America, and use this estimate to approximate the global richness of this ubiquitous and important parasite group. Collectively, these analyses suggest that there is still tremendous potential for biodiversity discovery of avian haemosporidians, with global avian haemosporidian richness possibly exceeding that of birds.

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Gregarina lutescens infecting the Harlequin Ladybird *Harmonia axyridis* (Coleoptera: Coccinellidae)

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Gregarina lutescens was described from the alimentary canal of the Harlequin Ladybird or Multicolored Asian Lady Beetle, *Harmonia axyridis* (Coleoptera: Coccinellidae) collected from Prairie Fleabane, *Erigeron strigosus*, at Peru State College, Peru, Nemaha Co., Nebraska. Our specimens differ from all 11 known species of *Gregarina* infecting coccinellid beetles worldwide by differences in size and relative shape, color, and association structure. *Gregarina lutescens* is smaller than 7 of the known species infecting coccinellid beetles but larger than the other 4 known species based on confidence interval exclusion of means. Our specimens are unique among known species of interest in their quince-yellow colored cytoplasm and precocious but ephemeral serial associations of up to 5 satellites. Nucleotide sequence (*18S*) phylogenetic analyses place the new species basal to a member of an internal clade of *Gregarina* that comprises gregarines parasitizing chrysomelid beetles. Phylogenetically, the analysis recovered 3 major lineages within the gregarines, representing the superfamilies Gregarinoidea, Stenophoroidea, and Stylocephaloidea, and demonstrating the propensity of gregarines to track host lineages and environments through evolutionary time. It confirms the polyphyletic nature of *Gregarina*, which currently comprises over 300 described species, only a handful of which are documented by genetic sequence suitable for phylogenetic analysis. Recollection, redescription, and molecular clarification of gregarine species infecting coccinellids would likely recognize a unique clade that would be an excellent system for studying the effect of intraguild host competition on parasite diversification and community structure. Ecologically, patterns of prevalence in this study indicate that *G. lutescens* reproduces primarily in larval hosts but depends on infections in adult beetles to overwinter, reflecting the differential vagility and frost tolerance of larval and adult host life cycle stages.

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Phylogenetic and morphological relationships from *Trichuris* (Nematoda: Trichuridae) parasites of Rodents in the Americas reveal independent lineages

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Trichuris is a cosmopolitan genus of mammal-dwelling parasites with 110 recognized species. There are three species with medical and economic importance: *Trichuris trichiura* (etiological agent of trichuriasis), *T. suis* (zoonotic trichuriasis), and *T. muris* (a model organism in parasitology). In stark contrast with their importance, there is no phylogeny nor systematic rearrangement for the taxon. In order to address this problem, we propose grouping species clusters defined by morphological characters to refine sampling for molecular attributes. The analyses revealed 12 clusters in *Trichuris* and 19 species that cannot be grouped based on deficient data availability. For this study, we used 20 specimens representing 10 species from the Americas as representatives of three of these clusters. The phylogenetic reconstruction based on the analysis of five genes reveals three new lineages in *Trichuris* comprehending 1- North American cricetids. 2.- South American hystricognath rodents (Cuniculidae, Dasyproctidae, and Ctenomyidae), and 3.-The North American group of Castorimorpha (Geomyidae, Heteromyidae). Species of *Trichuris* in North American Cricetidae form a monophyletic group with parasites of murids from Europe, suggesting relationships linking the North American and Eurasian Cricetidae (voles and woodrats) with Muridae (domestic mice); this group is characterized by the possession of medium size spicules (1.20-1.70 mm) and non-protrusible vulva. The Trichurids of North American castorimorphs form a tight monophyletic group occurring in pocket gophers (*Thomomys* spp.), kangaroo rats (*Dipodomys* spp.) and spiny pocket mice (*Heteromys* spp.), this clade is characterized by a cylindrical spicular sheath with uniform acute-saccular spines, and non protrusible to slightly protrusible vulvas. Finally, the South American hystricognath clade is represented by species with relatively long spicules, yet they feature great variability of other characters; this clade features significant divergence. The results suggest that the clades, including parasites of castorimorpha, hystricognaths, and cricetids, do not share an immediate common ancestor. The previous identification of clusters and sampling for molecular attributes resulted in a good strategy for reconstructing the phylogenetic relationships more efficiently. However, representative taxa from unsampled clusters are necessary to increase the resolution of the phylogenetic relationships in *Trichuris*.

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Marine Diseases: an overview

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Marine parasites fall outside the medical and veterinary traditions that dominate traditional parasitology courses. But parasites love the ocean. Along with their viral and microbial counterparts, they are a natural and diverse part of all marine ecosystems, coming from a wide range of taxonomic backgrounds that often differs from those on land. In part, this is due to differences in host taxonomies between land and sea. In most cases, marine parasites are simply doing their business unnoticed, just like any other creature. But in some cases, their effects are outsized, and they cause disease in marine life that we care about. This is particularly the case for fisheries and aquaculture. Although marine diseases seem to be declining in fisheries, they are increasing in aquaculture, which creates ideal conditions for parasite transmission. Often, it is wild parasites that plague aquaculture. But sometimes parasites from aquaculture escape into the wild and can wreak havoc. This talk may have you reconsider eating the sashimi at the buffet, but it is probably safer than crossing the street to grab a burger.

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Parasite life history interacts with large scale drivers of parasite abundance in Pacific marine ecosystems

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Though parasites are ubiquitous in marine ecosystems, determinants of parasite community structure are difficult to ascertain, with multiple potential drivers of the abundance and diversity of parasites interacting to shape the community. Marine parasites often are considered as a uniform group within ecosystems despite their significant diversity. Using a model selection approach within a database of more than 1200 individual fish hosts and their parasites from 11 islands within the Pacific Line Islands archipelago, we reveal that geographic gradients, including island area and island isolation, emerged as the best predictors of parasite abundance. Additionally, large scale ocean currents significantly impact parasitic community structure within some, but not all, hosts. Parasite life history interacts with these multiple drivers to share the community of parasites found within these hosts. This novel analysis of a unique dataset indicates that parasite abundance in marine systems cannot be predicted precisely without accounting for the independent and interactive effects of each parasite's life history and environmental conditions.

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Euhaplorchis sp. effect on social behavior and familiarity of Gulf killifish (*Fundulus grandis*)

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Trophically transmitted parasites may manipulate their hosts' phenotype (e.g., behavior, physiology, morphology) to increase the likelihood of transmission to the definitive host. In fishes, stable social groups develop familiarity over time through repeated interactions among individuals, and social preferences are often developed due to familiarity. Consequently, fishes often shoal with familiar fishes, a behavior that is likely to be protective against predation. Parasites may alter fish social dynamics in two ways: whether by decreasing association with familiar individuals, thereby isolating infected fish and making them more susceptible to predation by definitive hosts; and/or by incentivizing uninfected individuals to avoid infected fish in their shoal. In the present study, we tested whether Gulf killifish *Fundulus grandis* experimentally infected with *Euhaplorchis* sp. exhibited altered social preferences based on familiarity. We used a choice test methodology, allowing focal fishes to choose to associate with familiar or unfamiliar conspecific fishes held in transparent enclosures. We assessed whether infection status affected how often a focal fish was observed within two body lengths of familiar vs. unfamiliar conspecifics every second for 6 minutes; we also assessed their mean distance from familiar or unfamiliar groups. We then examined how these responses varied in infected or uninfected focal fishes. Our results are discussed in the larger context of parasite effects on host populations in coastal and estuarine environments.

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Emergence of *Scaphanocephalus* infections in Caribbean reef fish: Evidence from museum samples and contemporary sampling

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Despite evidence that diseases of marine wildlife are increasing in frequency and severity, historical data on infections are often lacking for time scales beyond a few years. Archived samples of marine hosts from natural history collections offer a powerful tool for evaluating whether specific parasites are new or changing in prevalence and intensity. Here, we used vouchered fish collections from the southern Caribbean to investigate long-term (1905-2022) shifts in infections by the trematode *Scaphanocephalus* spp., which has recently been linked to observations of Black Spot Syndrome in reef fishes. Examination of 191 museum-preserved fishes from Curaçao and Bonaire revealed that *Scaphanocephalus* infections are not new, with the earliest detections from a parrotfish collected in 1920. Distinctive morphological and histological traits of the metacercariae facilitated differentiation from other trematode taxa encountered. However, *Scaphanocephalus* infections were rare among archival surgeonfish and parrotfish, with a prevalence of 4.7% and a maximum intensity of 9. Contemporary collections of 242 ocean surgeonfish and parrotfish (7 species) supported a 14-fold higher infection prevalence (68.5%) and a 33-fold higher average infection abundance (11.9; range: 1 to 180). Fish body size and geographic location (Bonaire > Curaçao) also positively influenced infection. These findings offer quantitative evidence that infection by *Scaphanocephalus* has increased substantially over the past century and underscore the value of biological repositories in the study of emerging parasites in marine ecosystems.

We emphasize the need for additional research to assess the likely drivers of infection, its consequences for ecologically important reef herbivores, and the spatial extent of its emergence.

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Expanding on expansus: A new species of *Scaphanocephalus* from North America and the Caribbean based on molecular and morphological data

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Members of the genus *Scaphanocephalus* mature in accipitrids, particularly osprey, *Pandion haliaetus*, with metacercaria causing Black Spot Syndrome in reef fishes. In most of the world, only the type species, *Scaphanocephalus expansus (*Creplin, 1842) has been reported. Recent molecular studies in the Western Atlantic, Mediterranean and Persian Gulf reveal multiple species of *Scaphanocephalus*, but have relied on 28S rDNA, mainly from metacercariae, which limits both morphological identification and resolution of closely related species. Here we combine nuclear rDNA with mitochondrial sequences from adult worms collected in osprey across North America and the Caribbean to describe species and elucidate life cycles in *Scaphanocephalus*. A new species described herein can be distinguished from S. expansus based on overall body shape and size. Phylogenetic analysis of the whole mitochondrial genome of *Scaphanocephalus* indicates a close relationship with *Cryptocotyle*. We conclude that at least three species of *Scaphanocephalus* are present in the Americas and two others are in the Old World. Specimens in the Americas have similar or identical 28S to those in the Mediterranean and Persian Gulf, but amphi-Atlantic species are unlikely in light of divergence in cytochrome c oxidase I and the lack of amphi-Atlantic avian and fish hosts. Our results provide insight into the geographic distribution and taxonomy of a little-studied trematode recently linked to an emerging pathology in ecologically important reef fishes.

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Changes in Transmission Rates Drive Seasonal Patterns of Shrimp Black Gill Disease Caused by the Parasitic Apostome Ciliate *Hyalophysa lynni*

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Parasite prevalence is the net result of processes that increase and decrease the number of infected individuals in a population. Quantification of these processes is a difficult but necessary procedure required to both inform mechanistic-based models of parasite prevalence and to develop management strategies. Here, we perform a series of manipulative field and laboratory experiments to disentangle the role of transmission, mortality, and recovery in seasonal prevalence of shrimp Black Gill (sBG) disease; a condition caused by the apostome ciliate *Hyalophysa lynni* impacting the Georgia coast. We investigated disease transmission rates through five independent experiments conducted seasonally from July 2019 – April 2020. For each experiment, we deployed shrimp that had been treated to reduce sBG infection inside predator protection cages into the water column of Wassaw estuary in

Southeastern Georgia where sBG is prevalent and monitored the infection rate over a 10-day period. In a second series of laboratory experiments, we measured mortality due to *H. lynni* infection in the absence of transmission by comparing the mortality rate of shrimp that had been treated to reduce *H. lynni* infection and wild shrimp with naturally high levels of infection over 8 days at two ecological relevant temperatures. Finally, we estimated the recovery rate of sBG by monitoring the *H. lynni* prevalence and intensity in wild collected shrimp with initially high levels of infection over 22 days of holding shrimp in laboratory conditions. Transmission rates were highest during the summer with 75-91% of shrimp acquiring infection, but declined substantially in the fall, winter, and spring (0-10% prevalence). In our lab experiments, we observed complete recovery from infection with little disease induced mortality. Together, this study suggests that the seasonal rise and fall of sBG is due to high transmission rates of the parasite in the summer months followed by gradual recovery when transmission rates are low.

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Do cercariae of trematode species found in different intertidal habitats show consistency in key morphological traits?

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The persistence of parasite communities in time and space is intricately tied to successful life cycle completion, thus parasite traits that enhance transmission play a critical role. The free-living cercariae that emerge from their first intermediate molluscan hosts are known to exhibit various behavioral and morphological traits that promote encounter with second intermediate hosts. Cercariae of trematode species found in comparable habitats could therefore face similar challenges when trying to find the same or analogous hosts. Consequently, these cercariae could be more alike than expected when it comes to morphological traits that provide a benefit in this context. Here, we considered trematodes utilizing snails within intertidal habitats as first intermediate hosts to look for commonalities in various aspects of morphotype. We gathered data from 43 papers that conducted field studies in intertidal habitats from 28 distinct locations (and more sublocations within these) representing 15 countries and 6 continents. These studies encompassed over 150 different species of trematode using 19 snail host species. We looked for patterns related to the occurrence of 12 traits such as eye spots or a forked tail after assigning the cercariae to one of 16 morphotypes (e.g., monostome, strigeid). Similarities and differences in cercariae traits among intertidal habitats, including those characterized by different climates from tropical to polar, will be discussed.

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Parasites make important and consistent contributions to temperate and tropical marine systems

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Only a handful of studies incorporate parasites on the same ecological footing as free-living organisms, largely restricting our understanding parasites in ecosystems to temperate estuaries on the Pacific coast of North America. Parasites make significant contributions to these systems but it is unclear if this is a quirk of estuaries or a general feature of parasitism. We systematically incorporated parasites into a quantitative food web for a novel system, the sand flats at Palmyra Atoll. To test the generality of the parasite contributions to ecosystem structure we compared the role of parasites in the tropical Palmyra food web to that of three temperate estuaries. In both systems, parasites make contributions to richness, abundance, and biomass comparable to free-living consumer groups, such as birds. Further, in

both systems parasites dominate network structure in ways that free-living consumers cannot. These results suggest that parasites make general and important contributions to ecosystem structure. Our understanding of food webs is incomplete without parasitism.

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Parasites and Public Service: Research Vignettes from the Florida Fish and Wildlife Research Institute.

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The Florida Fish and Wildlife Conservation Commission (FWC) is unusual among equivalent agencies in US states in that it has an entire division primarily focused on conducting and publishing peer-reviewed scientific research, and this includes the study of aquatic animal parasites and other pathogens. 'Government parasitologist' is an uncommon job option, but the position unofficially exists in FWC because an early career fish parasitologist founded an aquatic animal health program and then stayed through retirement building the agency's capacity from the bottom up. As will be illustrated here, the program meets an often-neglected public need and participates in synergistic collaborations with academic and other governmental entities to accomplish otherwise impracticable research objectives. Collaborative research conducted by the group has for example led to changes in water management practices in South Florida, corrected gonad histology misinterpretations affecting regulatory decisions for sport fisheries, and dramatically expanded available information on fish parasite biodiversity in Florida.

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The journey from planarian to multidisciplinarian

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You have to admit that parasitology is a niche skill, and morphological taxonomic parasitology doubly so. From a start in digenean taxonomy grew a career that has had more twists than an M. Night Shyamalan movie. How did this happen? How does our academic preparation in a specific discipline truly prepare us for the myriad twists and turns of life and career? Is it really the hard work, or the decisions we make, or is it mostly <gasp!> luck? I argue that parasitology is, in fact, great preparation for a multidisciplinary career, especially with the right teachers and a healthy dose of serendipity. This is, of course, a totally teleological argument, but it helps me sleep at night, and hey, maybe it will work for you too.

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Exploring parasite-induced alterations in sociality and behavior in estuarine fishes

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Parasites often modify host behavior to enhance their own survival and transmission. In California, the trematode *Euhaplorchis californiensis* has been shown to infect the brain and modify the behavior of its second intermediate hosts- killifishes- making them more susceptible to predation by fish-eating wading birds, the parasite's definitive hosts. In Florida, we've been exploring how individual and social behavior are altered in gulf killifish (*Fundulus grandis*) following controlled infection with a closely related parasite (*Euhaplorchis* sp. nov.) with a nearly identical life cycle. We examined how a suite of behavioral traits (sociality, anti-predator response, and personality) are altered in experimentally infected hosts, with the long-term goal of understanding how such behavioral changes scale up to population and community level in natural habitats. For example, we explored how infection alters individual personality traits, using behavioral assays for boldness (tendency to take risks) and exploration (willingness to explore novel environments). Our data support the hypothesis that infected individuals are bolder and more exploratory than uninfected individuals, which presumably makes them more susceptible to predation. We also detected significant effects of infection on fast-start escape response reaction times, speed, and acceleration in response to simulated predators. As these small-bodied fishes play a key role in estuarine food webs, parasite induced changes in behavior could have far-reaching consequences in these ecosystems and may present a model for generating hypotheses that can be tested in host-parasite systems from similar habitats around the world.

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Specialist within a specialist: investigating infection patterns of helminth communities in osprey across North America with an emphasis on *Scaphanocephalus* spp.

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Osprey are one of the most unique and best-studied raptors worldwide, yet there are relatively few studies of their parasites in North America. The long-distance migrations, evolutionary isolation, and exclusive fish diet of osprey all have the potential to make their helminth communities distinct from other raptors. To better understand the parasite assemblages within North American osprey, we collected gastrointestinal tracts from wildlife rehabilitation centers and museums in the USA and Caribbean to determine the composition, prevalence, and load of the helminth community. We also paid particular attention to trematodes in the genus *Scaphanocephalus*, which are osprey specialists and have recently been identified as the etiological agent of Black Spot Syndrome, a condition that causes dermal lesions in the skin and fins of keystone herbivorous fishes throughout the Caribbean. Whenever possible, we used a combination of morphological and molecular analysis to determine the identities of detected parasites. We dissected a total of 223 osprey collected across 12 US states and the island nation of Curacao over 8 years. Overall, most detected parasites were trematodes, including at least 10 different genera. Additionally, we detected 4 nematode, 1 cestode, and 1 acanthocephalan genera. We detected multiple species of *Scaphanocephalus* infections in 16.6% of birds ranging in intensity from 1 to 394 worms (from a male osprey in Florida). The majority of infections were detected among osprey from Florida (34/37) with other detections in Delaware, Virginia, and Montana. No records of *Scaphanocephalus* were derived from birds in states along the Pacific flyway. Results are discussed in the context of the migratory patterns of osprey across the Americas and records of infection within fish second intermediate hosts from the Caribbean. In-progress work on the species identities and infection rates of collected worms will provide additional insights into interactions between this unique host and its parasite fauna.

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Parasites of parasites: The diversity of polychaete-gregarine-metchnikovellid associations in the Pacific Northwest

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Gregarine apicomplexans are dynamic single-celled eukaryotes that parasitize the extracellular spaces of invertebrates (e.g., the intestines) and are particularly common in marine polychaete hosts. The feeding cells of gregarines can harbor their own intracellular parasites (i.e., hyperparasites), called metchnikovellids. Metchnikovellids are recognized as early branching microsporidian fungi, which form a diverse and ecologically successful group of parasites. Despite their significance in understanding microsporidia evolution, the diversity of metchnikovellids remains poorly understood due to their cryptic nature as hyperparasites of marine gregarines. Over a two-year period in British Columbia, Canada, we screened diverse groups of polychaetes for parasites and hyperparasites. This effort led to the discovery of more than 30 species of gregarines and metchnikovellids, greatly expanding current knowledge of their diversity. We characterized the morphology of these novel parasites using light and electron microscopy; we also inferred single-gene phylogenetic trees based on small subunit ribosomal rRNA sequences for larger taxonomic coverage and multigene phylogenetic trees using single-cell transcriptomic and genomic data for better resolution of deeper relationships. Our analyses of these morphological and molecular data allowed us to reconstruct the phylogenetic relationships among major groups, identify cases of host specificity, and provide evidence for codiversification in this unique host-parasite-hyperparasite system.

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Marine Parasites as Colonizers in Waiting: Insights from the Panama Canal as a Natural Experiment on Biological Invasions

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Marine parasites often interact with freshwater systems, commonly via euryhaline hosts in transitional environments, thereby establishing the potential for colonization and invasion events. Occasionally opportunities are created by engineered environments, which provide a known timeline for studying biological invasions. The Panama Canal with its recently expanded locks provides a natural experiment to examine colonization/invasion events in real time and test hypotheses about invasion dynamics. Ongoing studies on the parasites of two common marine fishes that invaded Gatun Lake through the Panama Canal, namely the Atlantic Needlefish (*Strongylura marina*) and the Snook (*Centropomus undecimalis*) provide the first insights into the colonization dynamics of their parasites. Differential infections of Needlefish by two monogeneans (*Ancryocephalus* sp. and Axinidae gen. sp.) in marine and freshwater environments suggest different salinity tolerances and potential for freshwater colonization. In comparison, high intensities of intestinal infection of Snook by immature, maturing, and gravid individuals of a putative marine cryptogonimid species in Gatun Lake suggests successful establishment in freshwater. Examination of native and non-native freshwater fishes (~ 24 species, 12 families) from tributaries of the Chagres River and inshore waters of Gatun Lake over a 14-year period have revealed no marine parasites, indicating that spill-over effects from marine introductions have not yet occurred. In addition to potential spillover effects, the parasites of marine/euryhaline fishes, once established in Gatun Lake, greatly increase their opportunities for interoceanic invasions across the Panama Canal.

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Did brain-infecting scuticociliates (Philisterida sp.) kill wild sharks in the northwestern Atlantic Ocean?

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Most scuticociliates are free-living and range in marine environments but several can infect fishes opportunistically. During August and September 2018, moribund smooth dogfish (*Mustelus canis*; Carcharhiniformes: Triakidae) were observed stranding and/or washing ashore dead on Coney Island and Brighton Beach, New York, USA. When returned to the water, moribund dogfish failed to right themselves, swam short distances, and settled to the bottom before stranding again soon thereafter. Eight of these dogfish were necropsied and sampled. Grossly, we observed softening of the olfactory lobe, hemorrhaging of the meninges, and opaque, bloody cerebral spinal fluid. A biopsy of brain from 2 smooth dogfish was wet-mounted and observed with a compound microscope equipped with differential interference contrast optical components, revealing an infection by a species of Philasterida Small, 1967 (Ciliophora: Scuticociliatida) in all sharks examined. Infected brain exhibited acute necrotizing meningoencephalitis with intralesional scuticociliates; with some fields of view comprising mainly scuticociliates. A 584 base pair fragment of the small subunit ribosomal rDNA (*18S* rDNA) of the parasite was identical to sequences ascribed to *Philasterides dicentrarchi* Dragesco, Dragesco, Coste, Gasc, Romestand, Raymond, & Bouix, 1995(Philisterida: Philasteridae) and *Miamiensis avidus* Thompson & Moewus, 1964 (Philisterida: Parauronematidae). Our results agree generally with a 2017 study reporting mortalities of leopard sharks (*Triakis semifasciata*; Triakidae) in San Francisco Bay attributed to *M. avidus* infecting the brain. Evidence of any single pathogen killing a wild shark in nature is rare and report worthy. Our results indicate that these sharks died because of this infection. If so, this would constitute the first report of an infectious disease killing a wild elasmobranch in the Atlantic Ocean.

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Different life history strategies in the transmission of *Hematodinium perezi* (Dinoflagellata: Syndiniales) from the blue crab (*Callinectes sapidus*)

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The order Syndiniales is comprised exclusively of parasites, which have a global distribution in marine environments. Most syndinid dinoflagellates are parasitoids, killing their host with the final release of dinospores into the water column. Although many syndinid dinoflagellates remain relatively uncharacterized, the ecology of *Hematodinium perezi* is comparatively well documented from blue crabs. One feature of sporogenesis in *H. perezi* is that it produces two different types of zoospores, macro- and micro-dinospores, and upon sporulation each crab releases only one type of spore. This phenomenon has been reported for other syndinid dinoflagellates but has never been studied due to the ephemeral nature of the spores. The sexual cycle of *H. perezi* has yet to be determined and the function of the dinospores remains unknown. We employed multiple approaches to address two main questions in the life history of this parasite. What is the function of each dinospore type and why does each crab only produce one type of dinospore? Methodologies were developed in the laboratory to generate large quantities of high quality dinospores. The ploidy of each stage was investigated with flow cytometry and compared with the ameboid trophont i.e., the vegetative stage, which was collected from the hemolymph of infected crabs. Laboratory transmission using only the micro-dinospores was successful, suggesting that the two dinospore types are not gametes. In addition, RNASeq was done for transcriptomic analysis of the different life history stages. There were clear differences in the transcriptomic profiles of each cell type. Lastly, microsatellite markers are being used to test whether

dinospores are produced by all of the different strains of parasites in hosts infected with multiple strains. Altogether, our findings indicate that *H. perezi* has a unique life history with an internal sexual cycle and at least two different infection strategies.

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Quantitative ethology of schistosome miracidia and other aquatic invertebrates

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Miracidia are the short-lived first larval stage of digenetic trematodes that seek and infect the first intermediate host. Schistosome miracidia, the primary lab model for studies describing the behavioral processes involved in host-seeking and infection, engage in three distinct behavioral phases, integrating environmental and host cues to select an ideal microhabitat, scan the microhabitat for potential hosts, and locate and penetrate a host. Miracidia host-seeking is a critical chokepoint in the schistosome life cycle that could be exploited in schistosomiasis control efforts, especially in areas where mass drug administration alone is unlikely to eliminate the disease as a public health problem. Much work has been performed to qualitatively describe these behavioral phases, but diverse experimental approaches, host and parasite sources, and technological capabilities have resulted in often contradictory conclusions about the details of these behaviors. Furthermore, each behavior (collectively comprising the ethogram) has not been quantitatively defined. We have designed, fabricated, and optimized a cutting-edge imaging and analytical toolkit for quantitatively describing miracidia behaviors at unprecedented scale and precision. Using an array of high-resolution cameras, bespoke laser-cut and 3D-printed behavioral arenas, and custom Python analytical pipelines run on a high-performance computer – the InVision system – we record medium (6-10 cm from host), short (<1 cm from host), and contact-based behaviors, which can then be linked to the original qualitative ethogram. We use this technology to describe the behaviors of miracidia originating from different definitive host tissue sources, in the presence or absence of snails or snail-conditioned water, and in varying environmental conditions. Finally, we show how the InVision system can be extended to a variety of aquatic invertebrates, including mosquito larvae.

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No detection bias between two sampling approaches for intermediate hosts infected with brainworm (*Parelaphostrongylus tenuis*)

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Ungulate populations of conservation concern may benefit from interventions that would reduce brainworm (*Parelaphostrongylus tenuis*) transmission. One strategy to reduce transmission is to prevent encounters between infected gastropods and ungulates, since ungulates are infected via the ingestion of gastropod intermediate hosts containing the L3 stage of brainworm. Devising and employing this strategy requires knowledge of where infected gastropods occur, but they are rarely observed in nature. Consequently, it is unknown if infected gastropods are most likely to be found on vegetation or in the leaf litter/soil. Several hypotheses have been proposed to explain low gastropod infection prevalence, including whether certain gastropod sampling methods are biased against infected individuals. If brainworm infection alters climbing behaviour, then infected gastropods may be under sampled with cardboard traps (which only collect non-climbing gastropods), compared to visual searches (which allow for the collection of both climbing and non-climbing gastropods). To compare these sampling

approaches, we surveyed gastropods at six forested sites bimonthly from June-October 2023 where brainworm is known to occur. Gastropods were estimated from transects consisting of 30 cardboard traps, and 10-minute visual searches of similarly sized quadrats were performed alongside five randomly selected cardboard traps. Although the prevalence of L3s in gastropods was similar between the cardboard traps and visual searches (0.5% (1/194) and 0.4% (1/223), respectively), cardboard traps were significantly more likely to yield individuals of host species for brainworm. During visual searches, 3.6% of gastropods were climbing vegetation, and none were infected. Results from field sampling suggest that cardboard traps are not biased against infected gastropods, and that brainworm does not affect gastropod climbing behaviour. Our field-based study suggests that ungulate hosts of brainworm are most likely becoming infected while foraging along the ground, which is where land management strategies should be employed to reduce transmission. However, since so few individuals were found climbing and infected in nature, lab-exposures could help to further characterize the effect of infection on climbing behaviour. Results of ongoing lab experiments with the meadow slug, *Deroceras laeve*, will be discussed.

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It's a worm-eat-worm world: Consumption of trematode (Digenea) free-living stages protects snail hosts and benefits annelid predators

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Predation on parasites is a common interaction with multiple, concurrent outcomes. Free-living stages of parasites can comprise a large portion of some predators' diets and may be important resources for population growth. Predation can also reduce the density of infectious agents in an ecosystem, with resultant decreases in infection rates. While predator–parasite interactions likely vary with parasite transmission strategy, few studies have examined how variation in transmission mode influences contact rates with predators and the associated changes in consumption risk. To understand how transmission mode mediates predator–parasite interactions, we examined associations between an oligochaete predator (*Chaetogaster limnaei*) that lives commensally on freshwater snails and nine trematode taxa that infect snails. *Chaetogaster* is hypothesized to consume active (i.e., mobile), free-living stages of trematodes that infect snails (miracidia), but not the passive infectious stages (eggs); it could thus differentially affect transmission and infection prevalence of parasites, including those with medical or veterinary importance. Alternatively, when infection does occur, *Chaetogaster* can consume and respond numerically to free-living trematode stages released from infected snails (cercariae). These two processes lead to contrasting predictions about whether *Chaetogaster* and trematode infection of snails correlate negatively ('protective predation') or positively ('predator augmentation'). Here, we tested how parasite transmission mode affected *Chaetogaster*–trematode relationships using data from 20,759 snails collected across four years from natural ponds in California. Based on generalized linear mixed modeling, snails with more *Chaetogaster* were less likely to be infected by trematodes that rely on active transmission. Conversely, infections by trematodes with passive infectious stages were positively associated with per-snail *Chaetogaster* abundance. Our results suggest that trematode transmission mode mediates the net outcome of predation on parasites. For trematodes with active infectious stages, predatory *Chaetogaster* limited the risk of snail infection and its subsequent pathology (i.e., castration). For taxa with passive infectious stages, no such protective effect was observed. Rather, infected snails were associated with higher *Chaetogaster* abundance, likely owing to the resource

subsidy provided by cercariae. These findings highlight the ecological and epidemiological importance of predation on free-living stages while underscoring the influence of parasite life history in shaping such interactions.

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Digenean trematodes associated with hygrophylid snails at the Rio Grande Nature Center (NM) with special consideration of *Physella acuta* snails

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The distribution of digenean trematodes and snail host compatibility remain understudied across many different ecosystems throughout the globe. This study surveyed hygrophilid freshwater snails to assess digenean parasite diversity at the Rio Grande Nature Center, Albuquerque, NM. Field-collected snails were assessed for patent infections by shedding of cercaria. Morphological and molecular analyses (phylogenetic analysis of PCR-derived *28S* sequences) of strigeid-, echinostoma- and xiphidio-type cercariae from Physidae, Lymnaeidae, and Planorbidae revealed a diversity of digenean trematodes, including Diplostomoidae, Spirorchidae, Echinostomatidae, *Notocotylus* sp., and *Halipegus* sp. The collected samples were further investigated for suitability of *Physella acuta* as intermediate host for digenean trematodes. This snail species is comprised of a population (B) confined to North America (native range) and a globally invasive population (A). While *P. acuta* hosts a diverse array of trematodes in the native range, digenean trematode infections are rarely reported from *P. acuta* in invaded regions (i.e., population A snails). This could be due to more effective immunity of *P. acuta* population A versus population B. Alternatively, both populations have similar immunity, but compatible digenean parasites are lacking in the ranges invaded by population A *P. acuta* (Enemy Release Hypothesis). To address this question, the samples of infected *P. acuta* from the native range (harboring both A and B populations) were characterized by *cox1* haplotype to investigate infection prevalence for each population. Additionally, the baseline immune status of *P. acuta* snails from populations A and B, rewilded under field conditions was assessed by RNAseq and differential expression analysis. (We acknowledge the superintendents of the Rio Grande Nature Park, Albuquerque NM for field study permits.)

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Tenebrionid beetles shows attraction to some, but not all, acids found in the feces of rats infected with the tapeworm *Hymenolepis diminuta*

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It has been known for over 25 years that feces from rats infected with the tapeworm *Hymenolepis diminuta* are more attractive to the beetle intermediate host (Tenebrionidae) than feces from uninfected rats. Our prior research identified five volatile chemicals, small acids, present in infected rat feces but not in uninfected feces. Butyric acid was found in the greatest relative amount, but also found were 3-methylbutyric acid, 2-methylbutyric acid, pentanoic acid, and hexanoic acid. Limonene was shown to be present in uninfected feces but not in infected feces. We then studied the attraction of *Tenebrio molitor* and *Tribolium confusum* towards these acids. Trials consisted of 10 beetles, starved for 24 or 48 hours, placed in the middle of a petri dish arena, and offered water or ethanol (control) or one or more of the acids dried on clean glass microscope coverslips. We observed the beetles for 10

minutes and counted how many were near the control or acid at each minute. A related samples Wilcoxon signed rank test showed no significant attraction of both 24 (p=0.969) and 48 hour (p=1.0) starved *T. molitor* to a mixture of the 4 primary acids, but a significant (p=0.017) attraction to 3-methylbutyric acid and butyric acid (p=0.036). We also found a significant attraction of *T. confusum* to the 4-acid mixture when limonene was used as the control (p=0.01), though no significant attraction to individual acids. This study is the first to identify volatile chemicals from a tapeworm-infected host and show beetle attraction to particular acids that may enhance the likelihood of lifecycle completion.

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Life Cycle Variation and Host-Parasite Interactions of Four Congeneric Species of Turtle Acanthocephalans

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Turtle acanthocephalan life cycles are complex and thought to involve an ostracod intermediate host and a turtle definitive host. One species, *Neoechinorhynchus emydis*, is known to use an additional snail paratenic host in its life cycle. However, how snails become infected and whether other species of turtle acanthocephalans can infect snails remains unclear. This study evaluated the survival, development, and host use of four species of turtle acanthocephalans (*Neoechinorhynchus chrysemydis*, *Neoechinorhynchus emydis*, *Neoechinorhynchus emyditoides*, and *Neoechinorhynchus pseudemydis*) in their intermediate, paratenic, and definitive hosts. Experimental infections were conducted in two species of ostracods (*Physocypria* sp. and *Cypridopsis* sp.) and the freshwater snail (*Planorbella* cf. *P. trivolvis*). Additionally, ionomes were generated for these acanthocephalans and their turtle definitive hosts to better understand the host use by these parasites. Upon ingestion by ostracods, eggs of all four species of acanthocephalans hatched in both ostracod species. However, no further acanthocephalan development occurred in *Cypridopsis* sp., whereas all four acanthocephalan species grew and developed to cystacanths in *Physocypria* sp. Of the four species of acanthocephalans, *N. emydis* was the only acanthocephalan species to survive and establish in freshwater snail hosts; whereas cystacanths of the other three species of acanthocephalans were digested after being ingested by snails. In contrast, when snails were exposed to eggs of all four acanthocephalan species, eggs were passed in the feces, resulting in no infections. Finally, the overall ionomic composition of adult acanthocephalans and their turtle hosts suggested that the hosts and parasites are sharing host resources. However, specific elemental differences indicated that turtle acanthocephalans can accumulate heavy metals greater than their hosts. Results of this study support our previous field observations that 1) *Physocypria* sp. ostracods serve as intermediate hosts for *N. chrysemydis*, *N. emydis*, *N. emyditoides*, and *N. pseudemydis*; 2) *N. emydis* uses snail paratenic hosts, while snails are a non-viable route of transmission to turtles for *N. chrysemydis*, *N. emyditoides*, and *N. pseudemydis*; and 3) as adults, these four acanthocephalan species utilize their turtle definitive hosts similarly in terms of ionomic composition.

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Different metazoan parasites, different transcriptomic responses, with new insights on parasitic castration by digenetic trematodes in the schistosome vector snail *Biomphalaria glabrata*

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Gastropods of the genus *Biomphalaria* (Planorbidae) serve as vectors for *Schistosoma mansoni*, the most common causative agent of human intestinal schistosomiasis. Using improved genomic resources, overviews of how *Biomphalaria* responds to *S. mansoni* and other parasites can provide unique insights into the reproductive, immune, and other systems of invertebrate hosts, and their responses to parasite challenges. With Illumina-based RNA-Seq, we compared the responses of iM line *B. glabrata* at 2, 8, and 40 days post-infection (dpi) with the digenetic trematodes, *S. mansoni* and *Echinostoma paraensei*, or *Daubaylia potomaca*, a nematode parasite of planorbid snails. Responses were compared to unexposed time-matched control snails. Each parasite provoked a distinctive response with a predominance of down-regulated snail genes at all time points following exposure to either trematode, and of up-regulated genes at 8 and especially 40dpi following nematode exposure. At 2 and 8dpi with either trematode, several snail genes associated with gametogenesis (particularly spermatogenesis) were down-regulated. Regarding the phenomenon of trematode-mediated parasitic castration, we define for the first time a complement of molluscan genes that are targeted, as early as 2dpi when trematode larvae are still small. Differential gene expression of snails with trematode infection at 40dpi, when snails were shedding cercariae, was unexpectedly modest and revealed down-regulation of genes involved in the production of egg mass proteins and peptide processing. Surprisingly, *D. potomaca* provoked up-regulation at 40dpi of many of the reproduction-related snail genes noted to be down-regulated at 2 and 8dpi following trematode infection. Happening at a time when *B. glabrata* began to succumb to *D. potomaca*, we hypothesize this response represents an unexpected form of fecundity compensation. We also document expression patterns for other *Biomphalaria* gene families, including fibrinogen domain-containing proteins (FReDs), C-type lectins, G-protein coupled receptors, biomphalysins, and protease and protease inhibitors. Our study is relevant in identifying several genes involved in reproduction that are targeted by parasites in the vector snail *B. glabrata* and that might be amenable to manipulation to minimize their ability to serve as vectors of schistosomes. Supported by NIH grants R37AI101438 (ESL) and RO1 AI170587 (S-MZ).

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Genome wide association study of an African snail vector of schistosomiasis identifies genes associated with resistance to infection by *Schistosoma mansoni*

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Schistosomes are blood flukes transmitted by freshwater snails. The disease schistosomiasis, afflicting hundreds of millions of people worldwide, can potentially be controlled by blocking transmission to snails. Most cases of intestinal schistosomiasis occur in sub-Saharan Africa, where they are caused by *Schistosoma mansoni* that are transmitted by the snail *Biomphalaria sudanica* and related *Biomphalaria* species. In contrast to the better-studied neotropical vector *B. glabrata*, there has been little genomic work on African snails due to the absence of genetic models and inaccessibility of snail isolates. The genetic basis of African snail-parasite interaction is therefore completely unknown. Identifying African snail genes that convey resistance to infection may facilitate ways to leverage gastropod immune mechanisms and disrupt the parasite's life cycle where it is most needed. To uncover such immunogenetic pathways, we have generated inbred lines and an annotated genome assembly of *B. sudanica*. We used this assembly (0.94 Gb, N50=1.1 Mb) to support a genome-wide association study (GWAS) using pools of infected (N=493) and uninfected (N=295) F1 offspring of wild-caught Kenyan *B. sudanica* originating from Lake Victoria. The GWAS and developed amplicon panel for validating GWAS hits identified several genomic loci strongly associated with schistosome resistance phenotypes, including genomic regions known to influence parasite resistance in *B. glabrata* as well as surrounding previously uncharacterized genes. Genes associated with parasite resistance GWAS variants tended to contain transmembrane domains, suggesting a potential role in pathogen recognition. These results provide a first glimpse into the innate immune system of the major vector *B. sudanica* that will help inform schistosomiasis control strategies and future studies aimed at predicting or manipulating the vector competence of the snail host.

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SSS-Slippery systematics of slithering snake worms

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The Cyathocotylidae Mühling, 1896 is a globally-distributed family of digeneans parasitic in a wide range of definitive hosts: birds, fish, mammals, and reptiles. Few studies have generated molecular data for members of this family, and only one from a cyathocotylid parasitic in snakes. A few genera are known to infect snakes, including *Gogatea* Lutz, 1935, *Szidatia* Dubois, 1938, and *Mesostephanoides* Dubois, 1951. Members of these genera are only known from Africa and Asia. These genera have a long, convoluted taxonomic history with the validity of *Szidatia* and *Mesostephanoides* often being questioned. In the present study, we examined the relationships between cyathocotylid genera with a focus on those from snakes. We collected adult cyathocotylids from the intestines of snakes in Vietnam and Australia. Morphological and molecular (sequencing of the 28S rDNA and *cox*1 mtDNA) studies revealed the presence of 2 new species from Vietnam and Australia. This is the first report of a cyathocotylid from snakes in Australia. We also re-evaluate the status of 3 genera of snake cyathocotylids using combined morphological and molecular data. Identities of some other recently published cyathocotylids are re-evaluated based on DNA sequences.

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Supplemental description of *Caballerotrema annulatum* (Diesing, 1850) Ostrowski de Núñez and Sattmann, 2002 (Digenea: Caballerotrematidae) from a new host (*Electrophorus* cf. *varii*) and locality (Amazon River, Colombia) with phylogenetic analysis and emended generic diagnosis

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Herein, we provide a supplemental description of *Caballerotrema annulatum* (Diesing, 1850) Ostrowski de Núñez and Sattmann, 2002 (Digenea: Caballerotrematidae) based on specimens collected from the intestine of an electric eel, *Electrophorus* cf. *varii* (Gymnotiformes: Gymnotidae) captured in the Amazon River (Colombia). This caballerotrematid can be differentiated from its congeners by the combination of having the following morphological features: body surface spines forming contiguous transverse rows, concentric (wrapping dorso-ventrally around body), distributing into posterior body half (vs. restricted to anterior body half in *Caballerotrema brasiliense* Prudhoe, 1960; indeterminate for *Caballerotrema aruanense* Thatcher, 1980, and *Caballerotrema piscicola* [Stunkard, 1960] Kostandinova and Gibson, 2001); head collar lacking projections (vs. having them in *C. brasiliense*, *C. aruanense*, *C. piscicola*), narrow (head collar more narrow than maximum body width vs. the head collar being wider than the body in *C. brasiliense*, *C. aruanense*, *C. piscicola*); corner spines clustered (vs. corner spines distributing as two separated pairs in *C. brasiliense*, *C. aruanense*, *C. piscicola*); pharynx at level of the corner spines (vs. pharynx far anterior to corner spines in *C. brasiliense*, *C. aruanense*, *C. piscicola*); and testes ovoid

and non-overlapping (*C. aruanense*; vs. sinuous and overlapping in *C. brasiliense* and *C. piscicola*). Based on our results, we revise the diagnosis of *Caballerotrema* Prudhoe, 1960 to include features associated with the shape and distribution of body surface spines, orientation and position of head collar spines, cirrus sac, seminal vesicle, oviduct, Laurer's canal, oötype, vitellarium, and transverse vitelline ducts. We performed Bayesian Inference analyses using the partial large subunit ribosomal (*28S*) DNA gene to determine the affinity of our specimens among Echinostomatoidea. Our *28S* sequence of *C. annulatum* was recovered sister to that of *Caballerotrema* sp. (which is the only other caballerotrematid sequence available in GenBank) from an arapaima, *Arapaima gigas* (Schinz, 1822) (Osteoglossiformes: Arapaimidae) in the Peruvian Amazon. Our sequence of *C. annulatum* comprises the only caballerotrematid sequenced tethered to a morphological description and voucher specimens in a lending museum. The present study is a new host record and new locality record for *C. annulatum*.

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Gut secrets of raccoons: Unveiling enigmatic new species of Alaria

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Alaria is a relatively small genus of diplostomids (Digenea: Diplostomidae) that infect the intestines of mammal definitive hosts. The life cycle of *Alaria* spp. require amphibian second intermediate hosts. However, snakes and small mammals are frequently used as paratenic hosts, and humans may serve as accidental paratenic hosts. Pathology in paratenic hosts occurs due to the parasite's unusual mesocercarial stage. Recent study has demonstrated *Pharyngostomoides* and *Parallelorchis*, two diplostomid genera, to be synonyms of *Alaria*. Of the 11 nominal *Alaria* spp., five are known to infect raccoons. Four of the former *Pharyngostomoides* and *Parallelorchis* spp. are exclusively known from raccoons (*Procyon lotor*). The only other *Alaria* species reported from raccoons is *Alaria alata*, the type-species of the genus. However, *A. alata* has only been reported from crab-eating raccoon (*Procyon cancrivorus*) in Brazil, and likely represents a different species. In the present study, we collected representatives of several digenean taxa from raccoons in Georgia, USA which were included numerous *Alaria* spp. individuals. We utilized a combination of 28S and ITS rDNA and *cox*1 mtDNA sequences to explore phylogenetic relationships among members of the genus. Morphological and molecular study revealed the presence of a new *Alaria* sp. in Georgia, which is morphologically similar to the former *Pharyngostomoides* spp. Our materials also allowed us to re-evaluate a variety of *Alaria* spp. currently deposited in museums, some with associated DNA sequences. Our study revealed the presence of at least one other undescribed species from raccoons in Minnesota, and redescribed *Alaria adenocephala*. Our study demonstrates that even well-studied hosts in the USA may harbor currently unknown digeneans.

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A deeper look at hooks: Inter-relationships among neoechinorhynchid acanthocephalans

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Acanthocephalans are integral parts of ecosystems and can damage host populations. There is, however, an existing knowledge gap about most species of acanthocephalans and many of them have yet to be analyzed using modern technology, including DNA sequencing. The key objective of this project is to increase our understanding of features of Family Neochinorhynchidae, a diverse group of ~150 acanthocephalans of fish and turtles that consists of 18 genera worldwide. The approach is to compare species of the 9 genera that occur in the United States using morphological data, with a focus on hook morphology, and DNA sequence data. The first phase of this project and the focus of this presentation is the survey work to obtain representatives of species of each of these 9 genera from at or near their type localities. As a result of various surveys, we now have study sets of specimens of 8 of the 9 genera; a representative of the 9th genus will be obtained later this year. The samples acquired include *Tanaorhamphus longirostris* (Van Cleave, 1913) Van Cleave 1919 and *Gracilisentis gracilisentis* (Van Cleave, 1913) Van Cleave 1919 from Gizzard shad (*Dorosoma cepedianum*) from the Illinois River near Havana, Illinois; *Octospiniferoides chandleri* Bullock, 1957 from Eastern mosquitofish (*Gambusia holbrooki*) from the Florida Everglades; *Floridosentis elongatus* Ward, 1953 from White mullet (*Mugil curema*) from coastal Florida; *Atactorhynchus verecundus* Chandler, 1935 from Bolivar Peninsula, Texas; *Paulisentis missouriensis* Keppner, 1974 from southeastern Nebraska; *Octospinifer macilentus* Van Cleave, 1919 from localities in New York; various species of *Neoechinorhynchus* Stiles and Hassall, 1905 from various localities. This talk focuses on the specific results of field sampling and preliminary observations of the variable hook morphology represented by these genera.

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Two and a half centuries of morphological mishaps within *Axine* Abildgaard, 1794 (Monogenoidea: Axinidae Monticelli, 1903): new observations of type specimens, description of a putative innominate species, and (finally) a tree that includes an axinid sequence

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Species of Axinidae Monticelli, 1903 infect the gill epithelium of mainly flyingfishes (Exocoetidae), halfbeaks (Hemiramphidae), and needlefishes (Belonidae) (all Beloniformes). We collected specimens representing a putative innominate species of *Axine* Abildgaard, 1794 from 2 Atlantic flyingfish, *Cheilopogon melanurus* (Valenciennes, 1847) (Beloniformes: Exocoetidae) from the outer continental shelf, northern Gulf of Mexico. Specimens were fixed properly for morphology and preserved in 95% EtOH for sequencing the large subunit ribosomal (*28S*) gene. A review of the taxonomic literature on *Axine* spp. (1794–2023) reveals that previous authors unanimously mistook the female accessory canal (perhaps responsible for spermatophore acceptance and maintenance) and its associated conical sclerite as comprising the vaginal pore and duct. We studied extant type specimens and our newly-collected specimens using differential interference contrast (DIC) optical components. We concluded that the vaginal duct of *Axine* spp. is a duct separated from the accessory duct and that is subdivided into a distal (near vaginal pore) and a proximal portion demarcated by an obvious constriction. The distal portion of the vagina comprises a spermatophore chamber (all of our specimens had a spermatophore) as well as a follicular chamber. The proximal portion of the vagina comprises a thick-walled and glandular chamber connected to the narrow uterine duct. Our specimens differed from all congeners (except *Axine belones* Abildgaard, 1794 and *Axine depauperati* Yamaguti, 1968) by having 35–40 testes. They differed from the published descriptions of *A. belones* by having a relatively transverse (vs. longitudinal) accessory piece and by lacking a markedly bifid vaginal sclerite. They differed from that of *A. depauperati* by having a body 4.8–6.0 × longer than wide and 62–81 haptoral clamps (vs. 13.6–13.8 × and 22–31, respectively). The *28S* phylogenetic analysis recovered the putative innominate species in a clade of other Mazocraeidea spp. It was sister to species of Heteromicrocotylidae Unnithan, 1961, Heteraxinidae Unnithan, 1957, and Microcotylidae Taschenberg, 1879. All of these taxa were monophyletic and sister to species of Diplozoidae Palombi, 1949. This is the first axinid collected and described from a flyingfish in the western Atlantic Ocean and first published nucleotide sequence for any axinid.

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Coccidial descriptions in the molecular age – with a focus on *Eimeria* species

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Advancements allowing the molecular characterization of coccidia have provided new opportunities for describing and differentiating species with more accuracy than ever before; despite this, the lack of publicly available sequences ensures that species comparisons continue to rely mainly on morphological descriptions. Unfortunately, there are many examples of species characterizations that only include oocyst morphology and morphometrics; overlapping dimensions and features of oocysts further complicate differentiation. Ideally, modern descriptions and redescriptions of coccidia should consist of complementary molecular and biological data including complete endogenous development characterized in short intervals (e.g., every 6 hours) whenever possible. Examples from our recent descriptions of new *Eimeria* species illustrate how detailed analysis of endogenous stages can facilitate differentiation of species but also provide crucial information on how each coccidium parasitizes its host. Lack of such data leaves many unanswered questions that seriously limit our ability to erect a stable coccidian taxonomy. For example, despite being considered enteric parasites, development of some *Eimeria* species is not limited to infections of enterocytes and instead takes advantage of intraepithelial lymphocytes for the transport of sporozoites and, in some cases, development of first-generation merogonic stages. In cross-transmission studies with some of these *Eimeria* species, it has been demonstrated that some infect multiple hosts, contradicting the assumption that *Eimeria* species and closely related coccidia are largely host-species specific. In a phylogenetic analysis based on mitochondrial cytochrome *c* oxidase subunit I sequences, *Eimeria* species that infect the same avian host are not necessarily the closest genetically to one another; instead, parasites infecting the same region of the intestinal tract tend to be monophyletic. Worse, monophyly of the genera *Eimeria* and *Isospora* is called into question in the same analysis. These exceptions in the biology and phylogeny of these parasites complicate assignment of any new species to the genus *Eimeria* and inference of relationships among such species. Describing new parasites and updating incomplete species descriptions using up-to-date methods is necessary to better understand these exceptions. These new, more comprehensive, observations may raise questions regarding the definition of coccidian genera and may necessitate that we revisit and revise our definitions for them.

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Purchasing your first pet coccidian: the diversity of coccidia and related parasites (apicomplexa) from pet store squamate reptiles

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Coccidia are a group of obligatory intracellular parasites that infect all classes of vertebrates. However, coccidia of reptiles are an understudied group. For example, there are approximately 6,300 species of lizards in 40 families, but only a few hundred species from 15 lizard families have been evaluated for coccidia infections. However, it is estimated that the coccidian diversity in lizards is extremely rich, with estimates suggesting as many as 12,600 species. More importantly, many of the coccidian species descriptions from lizards were based on oocyst morphology. However, recent studies indicate that to resolve the taxonomical and phylogenetic issues of coccidian parasites, it is critical to combine morphological studies of oocysts with descriptions of their endogenous development along with obtaining sequence data. Surprisingly many of the coccidian species descriptions from lizard hosts came from lizard species that are now maintained in zoos or breeding facilities for the pet industry. For

example, of the 19 common lizard species available for purchase from two large pet store chains in the US, 13 species of lizards have had 29 species of coccidia described from them. However, only 5 and 2 of those coccidian species descriptions have complimentary histological and/or sequence data associated with oocyst morphology, respectively. In this study, we sampled feces from squamate reptiles from pet stores in Oklahoma and evaluated them for coccidia and related protozoan infections for future molecular and endogenous studies on these parasites. We found that of the 5 species of squamate reptiles sampled, 3 species were infected with 2 genera and at least 3 species of coccidia and/or related protozoans. These included *Cryptosporidium* sp. and *Isospora jaracimrmani* from the Yemen chameleon, *Chamaeleo calyptratus*; *Cryptosporidium* sp. from the red tegu, *Salvator rufescens*; and *Isospora amphiboluri* and *Cryptosporidium* sp. from the bearded dragon, *Pogona vitticeps*. Our study indicates that coccidia infections are common in pet store reptiles. More importantly these infections provide an untaped resource for obtaining additional sequence and endogenous developmental data to complement previous descriptions which were based strictly on oocyst morphology. New information on oocyst morphology and sequence data of reptilian coccidia will be discussed.

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Diversity of *Quadriacanthus* Paperna, 1961 (Monogenea: Dactylogyridae) in the Upper Congo Basin: new geographical records and description of five new species from the gills of *Clarais ngamensis* Castelnau, 1861 (Siluriformes: Clariidae)

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Monogenean parasites belonging to *Quadriacanthus* have never been reported on the blunt-toothed African catfish *Clarias ngamensis* but are commonly found on congeners. A parasitological survey of this fish species can lead to the record of multiple monogenean species, potentially new for science. The present study aims to explore the monogenean parasites belonging to *Quadriacanthus* from the gills of *C. ngamensis* in the Upper Congo Basin by (i) inventorying their diversity and providing their morphological characterisation, and (ii) analysing their infection parameters. *Clarias ngamensis* was selected for the study, given its presence and economic value in the Upper Congo Basin. Monogeneans were isolated from the gills, mounted on glass slides with ammonium picrate-glycerin for identification based on morphological analysis of slerotised parts of the genitalia and attachment organ. Species richness was reported and infections parameters of the retrieved parasites were calculated. Potential mechanisms of diversification of *Quadriacanthus* spp. and inference of the genetic proximity of *C. ngamensis* and *C. gariepinus* on the basis of the parasitic species they share, are also discussed. Seven parasites species were found, among them *Q. aegypticus* El-Naggar & Serag, 1986, *Q. allobychowskiella* Paperna, 1979 were recorded on *C. ngamensis* as new host. The Upper Congo Basin is a new geographical location for them, as well for the five other species newly described. Prevalences of all parasite species range 7 – 67% and mean intensities 1 – 6 parasites per infected fish. Since *Quadriacanthus allobychowskiella* and *Q. aegypticus* are known to date as parasites of *C. gariepinus,* their presence on *C. ngamensis* is proposed to result from a possible host switching. This record of seven species of *Quadriacanthus* is consistent with the hypothesis that the potential host diversity in the tropics and the narrow specificity of *Quadriacanthus* spp. could lead to the discovery of several, even new, parasite species, and it highlights the potential biodiversity still to be explored in the Upper Congo Basin.

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Montane molluscs: factors affecting snails and their trematodes throughout the mountain ecoregions of Arkansas

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Trematodes in the subclass Digenea are of both medical and veterinary importance. Digeneans require an obligate molluscan first intermediate host to complete their life cycle, highlighting the importance in understanding local host-parasite relationships. My research aims to (1) quantify trematode morphospecies prevalence among several snail taxa, (2) investigate and describe patterns of host-parasite species richness across lotic and lentic communities, and (3) describe which landscape factors affect snail and trematode occupancy. During the summer of 2022, we visited 44 sites (24 lotic, 20 lentic) in the Ozark Highlands and Boston Mountains ecoregions and individually monitored 6,934 snails for trematode infections. The caenogastropod snail species *Pleurocera canaliculata* had the highest trematode prevalence at 15.8%. During the summer of 2023, we visited 64 sites (34 lotic, 30 lentic) in the Ouachita Mountains ecoregion and assessed 4,143 individuals for trematode infections. The hygrophilan snail species *Helisoma anceps* had the highest trematode prevalence of 7.8%. Overall, 13 cercarial morphotypes were identified from the Ozark ecoregion and 11 morphotypes from the Ouachita ecoregion. We sequenced representative snails using genes 28S and 16S and the cercariae they shed using the genes 28S and 18S, which is currently being analyzed. The datasets that have been generated from this monumental effort represents one of the largest co-datasets on snail-trematode ecology and genetics.

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Testing evolutionary theory intersecting complex life cycles and hermaphroditic mating systems from field-collected parasites

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The mating system model of Brown et al. (2001) argues that complex life cycles concentrate trophically transmitted parasites into hosts further up the food chain, thus enabling more outcrossing opportunities (i.e., the model assumes complex life cycles reduce chances for inbreeding). The model also predicts that life cycle truncation can only occur when selfed offspring have the same or greater fitness as outcrossed offspring (i.e., there is no inbreeding depression). We provide the first explicit test this assumption and prediction. Our study system was the hermaphroditic trematode *Alloglossidium renale*, which has a truncated 2-host life-cycle, with sexual reproduction occurring within the paired antennal glands of grass shrimp. We first evaluated the mating system of *A. renale* from 3 different populations (Louisiana, Texas, and Mississippi) with a temporal replication in Mississippi (2014 and 2018). We next provided a novel test for inbreeding depression from field-collected samples. Specifically, we compared genetic estimates of selfing with demographic estimates of selfing. The premise behind the method is that if a demographic estimate of selfing (derived from the inverse of infection intensities, i.e., there is random mating within antennal glands) is greater than a genetic estimate, then fewer selfed offspring are surviving to adulthood relative to outcrossed offspring. Our results showed very high genetic estimates of selfing across all 4 samples (71.1-84.4%). The demographic to genetic selfing-rate comparisons provided no support for inbreeding depression. In fact, demographic estimates that incorporated density-dependent fecundity, could fully explain the high selfing rates. Our results support

the assumption and prediction of the Brown et al. (2001) model and demonstrate how demography (i.e., the distribution of infection intensities) can shape parasite mating systems.

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Changes in environmental conditions determine a bizarre host manipulation in Canada's fluke-infected zombie ants

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Parasitologists have yet to fully understand why parasites alter their host's behavior. The question of 'how' parasites orchestrate radical changes in host behaviors is even more vexing. I study fluke-infected zombie ants (*Dicrocoelium dendriticum*-infected Formicid ants) in a parkland region in Alberta, Canada, where the parasite has been introduced from Europe. In this example of host behavior manipulation, ants infected with a single brainworm attach their mandibles to a plant for hours or days. In a departure from classic cases of parasite manipulation, the ant does not die on the plant. Rather, it detaches from the plant to resume normal ant activities, then attaches again. This attachment/detachment sequence likely facilitates transmission into obligate grazing mammals. It is hypothesized that cues linked to attachment and detachment are closely linked to changes in temperature, but the empirical evidence available to support this hypothesis is limited. My field-based observations are designed to determine if the timing of attachment is associated with changes in environmental factors, alterations to the host's circadian rhythm, or both. I marked hundreds of ants on plants from infected nests with paint and followed the number of infected individuals on each plant for select consecutive days from May to August, 2023. Our results indicate that the number of attached ants varied throughout the day according to precise temperature thresholds. Despite this link with temperature, I also found a potential link between ant circadian rhythm and attachment time, with an increase in attachment at 6:00 p.m. every day, regardless of temperature. This work will characterize the patterns of manipulation in fluke-infected ants to provide the foundation for future mechanistic studies on this and other complex host manipulation systems.

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Following the snail trail: Uncoiling the factors that drive the distributions of trematodes among multiple ecoregions

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Our basic question is: why are trematodes located where they are? Snails play a fundamental role in parasite transmission of multiple phyla and are a keystone clade for freshwater trematodes. Despite snails being globally ubiquitous (excluding Antarctica), the abiotic and biotic factors that drive snail population and community dynamics remains mostly unknown, leading to fundamental data deficiencies on trematode community structure and co-evolutionary patterns, such as host specificity. Our research begins to address this gap by modeling the effects of local, landscape, and spatial factors on metacommunity structure, alpha diversity, and beta diversity of trematodes among hygrophilan and caenogastropod freshwater snails. In 2022 and 2023, my team surveyed 4,832 snails for trematodes among 44 sites in the Ozark Mountains and 4,859 snails among 65 sites in the Ouachita Mountains ecoregions of Arkansas. When we observed cercariae, we sequenced representative cercariae (18S & 28S) from that site and the snail host (16S & 28S) from which it was shed. We used an evolving metacommunity ecology framework to analyze the hierarchical structure of our data, ranging from a trematode species within a snail, trematode prevalence within a host species at a site, trematode community structure within the host snail community, and wetland & landscape factor effects on trematode community structure. Briefly, among the 101 sites and 9,691 snails surveyed, there were 199 (Ozarks) and 157 (Ouachitas) unique samples of hosts surveyed per site. Among those, 36% of the sampled host populations shed cercariae in the Ozarks and 57 (36.3%) of the sampled host populations shed cercariae in the Ouachitas. Our results have major implications for understanding the ecological and evolutionary processes that shape trematode community patterns in nature.

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Scratching at the phylogeny of the pocket gopher lice: assessment for *Geomys bursarius ozarkensis*

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Pocket gophers (family Geomyidae) and their chewing lice (genus *Geomydoecus*) are a model system for studying host-parasite codiversification because the lice are obligate parasites and are usually restricted to a single host species. Geographic isolation of host species into populations and subspecies is predicted to reinforce codiversification between the hosts and their obligate parasites. The Ozark pocket gopher, *Geomys bursarius ozarkensis*, is a highly isolated subspecies of the plains pocket gopher complex, only found in Izard County, AR. When initially described *G. b. ozarkensis* was assumed to share the chewing louse *Geomydoecus spickai* with the geographically closest subspecies of plains pocket gopher, *G. b. missouriensis*. We aimed to identify lice species of *G. b. ozarkensis* using morphological data from 15 male and 10 female lice, combined with molecular data (*cox1*) from 15 lice collected from 15 gophers. Based on morphology and genetics, we found that *G. b. ozarkensis* has a unique chewing louse species relative to other species in the complex. The louse was morphologically distinct based on the total body length, scape distal width, scape medial width, and spur length. The estimated phylogeny, based on *cox1* sequences, further indicated that the louse species was genetically distinct. This research reveals that barriers to contact between closely related host taxa may lead to speciation in their ectoparasites.

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Findings on *Myxobolus cerebralis* in newly invaded Alberta, Canada

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Since the initial discovery of whirling disease in Canada in 2016, efforts have concentrated on understanding and preventing the spread of *Myxobolus cerebralis*, largely due to its capability to cause severe population decline to salmonid fish in affected regions. Recent efforts have led to the development of an environmentally focused qPCR assay, which can be used to detect infection in *Tubifex tubifex* hosts, triactinomyxon actinospore filtrations, and environmental DNA (eDNA) filtrations. By focusing on environmental monitoring, we've been able to advance our understanding of the biology of *M. cerebralis* in Alberta. For example, we've found that a temperature range of 10-17°C is required for the proliferation of *M. cerebralis* within *T. tubifex*. This temperature range displayed an overlap with when juvenile salmonid fish are most susceptible to *M. cerebralis* infection in Southern Alberta. It may also serve as an environmental barrier to the further advancement of *M. cerebralis* into the colder waters of Northern Alberta. More recently, *T. tubifex* sampling has allowed us to carry out comprehensive phylogenetic analyses using completed mitochondrial genomes. This work has revealed that the formerly used lineage groupings are irrelevant within Alberta *T. tubifex* populations. A

comparison of the mitochondrial genomes of representative worms sampled from several populations of *T. tubifex* collected throughout Alberta suggests five unique sub-species exist. Two of these subspecies, T1 and T3, appear to be primarily responsible for *M. cerebralis* transmission in Alberta. These subspecies dominate rivers in Southern Alberta, and preliminary investigations of watersheds in the northern and central regions of the province are currently under examination to determine the risk of invasion. Our future work aims to complete a *M. cerebralis* challenge experiment of each subspecies to confirm these *T. tubifex* host specificity dynamics. Going forward, work on whirling disease in Alberta will continue to use eDNA to monitor the spread of the parasite. Additionally, the newly confirmed environmental and biological barriers such as oligochaete genetic information and river temperature can influence watershed management and fish stocking efforts within Alberta and British Columbia as *M. cerebralis* continues to spread.

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Determining the optimal density of cercariae for parasite oxylipin profiles

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Although signaling molecules serve as a potential mechanism mediating host-parasite interactions, including the modification of host behaviour, the precise roles of host and parasite signaling molecules remain elusive due to the obscured source of these molecules. Oxylipins, oxidized fatty acids, are signaling molecules produced by both hosts and parasites. Differences in oxylipin profiles between infected and uninfected hosts have been documented in several echinostome-snail species interactions. However, the specific contributions of parasites and hosts to these altered emission profiles are unknown. To address this gap, we employed lipid extraction techniques coupled with high-performance liquid chromatography/tandem mass spectrometry to characterize the signaling molecules from the trematode *Echinostoma trivolvis* lineage c. However, it was crucial to first determine the density of cercariae required for detection using our approach. We hypothesized that higher densities of cercariae would result in more diverse profiles of oxylipins being emitted at increased concentrations. Freshwater snails *Stagnicola elodes* were exposed to 5-10 miracidia, and 6 weeks later, these snails were placed under lights to assess for the emergence of cercariae. Subsequently, parasites from 13 infected snails were pooled and rinsed 3 times to eliminate host-derived molecules. The resulting pool was then subdivided into several samples, the density of cercariae in each sample was estimated, and water was conditioned with parasites for 4 hours. We obtained 6 water-conditioned samples with cercarial densities ranging from 91-158 cercariae/mL, and 2 negative controls. Following biochemical analysis, we will compare the diversity and amounts of oxylipins to determine the effect of density on oxylipin diversity and amount. These outcomes will aid in selecting an optimal density for the final sampling of cercarial oxylipins, allowing for the comparison of these signatures to previously published data from infected snails. Through these comparisons, we aim to understand the respective contributions of parasites and hosts to emissions from infected hosts. Additionally, determining the source of oxylipins will guide us in selecting oxylipins for assays aimed at elucidating their influence on parasite-modified host behaviour.

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Those who forget the past are condemned to repeat it: Evidence of repeated zoonotic pathogen spillover at ecological boundaries

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Anthropogenic modifications to the landscape have altered several ecological processes worldwide, creating new ecological boundaries at the human/wildlife interface. Outbreaks of zoonotic pathogens often occur at these ecological boundaries, but the mechanisms behind new emergences remain understudied. Here, we explore the roles of landscape use and biotic communities to provide a better mechanistic understanding of zoonotic disease emergence in transition zones. Using a dataset encompassing more than 65 species ranges and 480 presence-pseudoabsence localities in sub-Saharan Africa, we test the influence of macroecological drivers of diseases related to evolutionary strategies of host-disease interaction and landscape attributes that apply to any directly transmitted pathogen. We combine pathogen reservoirs and accidental host ranges of a deadly human pathogen, Ebolavirus, with landscape use in a machine learning framework. Our results show that overlapping species range edges and heightened habitat transformation increases Ebola outbreaks risk in ecological transition zones. Moreover, we provide evidence that several macroecological theories are intertwined to explain these patterns, with the potential for synergistic effects between mechanisms we consider. With increasing landscape changes worldwide, we provide novel ecological and evolutionary insights into our understanding of zoonotic pathogen emergence that highlights the risk of aggressively developing ecological boundaries.

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Tracking Extracellular Vesicle Release in Nematodes

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Parasitic nematodes are a global cause of disease in humans and animals. Nematodes emit extracellular vesicles (EVs) that interact with the cells of a host organism and interfere with their functioning in a way that is beneficial to the parasite. As cells can emit hundreds of EVs, quantifying by hand can be an inconsistent and time-consuming task, necessitating the use of unbiased software. CME Analysis is a free MATLAB program built to gather data on clathrin-mediated endocytosis and we are currently adapting CME Analysis to the needs of EV analysis. New MATLAB functions were compared to hand-counted reference images of EV emissions in *C. elegans* embryos. *C. elegans* are used as a non-parasitic model organism to study the cell biology of nematodes. CME analysis worked well with spatially distinct EVs but tended to undercount when EVs cluster. We have altered program settings and written additional scripts that have improved CME Analysis's ability to detect EVs in high emission strains and reduced undercounting. Add-ons to the program include the use of new MATLAB functions that take object size, coloration, and background noise into account. This new code will help create a more accurate count in areas of high EV density, and better differentiate objects from background noise. In the future, these assays will be applied to parasitic nematodes. Gathering accurate data on the EV emissions of parasitic worms can provide key insights into intraspecies communication and host-parasite interactions on a sub-cellular level.

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Costus afer and *Costus spectabilis* enhance survival and promote host immune response against *Plasmodium berghei* infection in mice

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To combat drug resistance in malaria and provide alternative treatment options, new antimalarial drugs, drug combinations, or alternative strategies are urgently needed. Plant-derived compounds have shown promise in effectively protecting against malaria infection by modulating the balance of pro-inflammatory (TNF-α) and anti-inflammatory (IL-10) cytokines. Hence, identifying potent antimalarial and immune-modifying compounds from plants could offer an alternate route to address drug resistance. Here, we investigated the antimalarial and immunomodulatory potential of *Costus afer* and *Costus spectabilis* in a mouse model of *Plasmodium berghei* infection. We investigated the antimalarial and immunomodulatory properties of solvent extracts derived from *C. afer* and *C. spectabilis* using a murine model of *P. berghei* infection. We assessed patent parasitemia, animal survival, and levels of pro- and anti-inflammatory cytokines through ELISA to determine the potential of the extracts in treating rodent malaria infections. The dose-dependent effect of methanol and hexane extracts of *C. spectabilis* and *C. afer* was observed in reducing parasitemia in a murine model of *P. berghei* infection. The highest concentrations of these extracts significantly ($P < 0.05$) inhibited parasitemia and significantly ($P < 0.05$) improved survival. Furthermore, the hexane extracts of both plants demonstrated a potent immunomodulatory effect by enhancing the production of both pro-inflammatory cytokine TNF-α and anti-inflammatory cytokine IL-10. The results of this study demonstrate that *C. afer* and *C. spectabilis* possess both antimalarial and immune-modulating properties against *P. berghei* infection. The results of our study suggest a need for further research to identify the specific molecules responsible for the observed cytocidal and immunomodulatory effects of these plants.

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wrmXpress GUI: A user-friendly interface for high-throughput analysis of parasitic worms

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As antiparasitic research moves towards high-content and high-throughput phenotypic screening, computation of large imaging datasets will play a significant role. The data generated by automated microscopy is invaluable to researchers as it is highly detailed and at an unprecedented scale. However, processing these data is a challenging task, requiring researchers to develop custom tools, use closed-source proprietary code integrated into the microscope’s software, or adapt publicly available open-source software. However, creating custom tools and working in the command line poses a barrier to entry for many researchers. With these considerations in mind, we have developed an open-source tool with a user-friendly GUI to facilitate the analysis and interpretation of high-content imaging data across diverse worm species. wrmXpress is an open-source tool designed to quickly analyze the data received from automated microscopy imaging with a special focus on parasitic worms. The new wrmXpress GUI incorporated all previously published modules, and we also introduce a new module for tracking schistosome miracidia in a high-throughput context.

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Sequencing snails and their trematode cercariae reveals host specificity patterns and cryptic diversity among multiple ecoregions

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Digenean trematodes are a diverse clade of parasitic flatworms that require their intermediate gastropod hosts to complete their life cycle. In many cases, the snail-trematode relationship shows apparently high host specificity, however, this needs to be investigated further given the high amount of cryptic diversity that likely exists among several life cycle stages of trematodes. In this study, we used sequences from trematode cercariae to create a phylogeny and assess host specificity and cryptic diversity. To survey for trematodes, we sampled thousands of snails in 2021 and 2022 among two ecoregions of Arkansas. The most common snails were in the families Physidae, Lymnaeidae, Planorbidae, and Pleuroceridae. Cercariae were identified to morphotype and then both the co-associated host and cercaria were sequenced. Our results focus on the analysis of the 28S and 18S ribosomal DNA sequences from trematodes. In total 40 trematode genera were identified in the 120 infected snails that were also sequenced. Snails in the family Planorbidae had the most trematode taxa, with Physidae having a close second. Our preliminary analyses revealed host specificity patterns among some trematode taxa and cryptic diversity. The results of this research represent one of the largest sequencing datasets for co-associated snails and trematodes.

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Comparative Preservation Methods for Parasites in Fecal Samples: Vacuum Sealing vs. Conventional Storage

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Fecal egg floatation is the conventional method for detecting many parasitic diseases. While this technique is considered the gold standard by wildlife researchers and veterinarians, there is significant variation in the preparation and storage of fecal samples before processing. The recommended processing time for fecal samples is within 48 hours. However, this is not always possible due to time constraints in the lab and field. Delays in processing can result in the development of parasite eggs, reducing the accuracy and precision of counts. Understanding how sample preparation and storage affect egg counts is important for interpreting results. While fecal preparation and storage methodologies have been investigated for domestic horse infections, these approaches have not been studied in wild horses which may harbor unique parasite communities. In this study, we compared the precision of fecal egg counts in samples stored in vacuum-sealed and ziplock bags over a 30-day period. The egg counts did not change regardless of treatment group. This indicates that both storage treatments are sufficient for sample preservation for wild horse parasite quantification. These results differ from previous studies on domestic horses and potentially lengthen the period between sample collection and processing.

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Parasite communities in sunfish (Centrarchidae) from the Olentangy River, Ohio, USA

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A parasite survey of sunfishes was carried out at three sites in the Olentangy River, Delaware, Ohio. A total of 413 fish were collected from August 2022 through December 2023 and examined for parasites. We found 22 parasite species in the centrarchid population. Metacercariae (mostly *Posthodiplostomum minimum*) and *Spinitectus* spp. were the most common parasites recovered. Additional parasites included 3 species of acanthocephalans, 2 cestodes, 3 nematodes, 7 trematodes, monogeneans, crustaceans, and 3 species of leeches. Bluegill (*Lepomis macrochirus*) had a larger abundance of parasites than green sunfish (*Lepomis cyanellus*). Other species including rock bass (*Ambloplites rupestris*), orange-spotted sunfish (*Lepomis humilis*), northern sunfish (*Lepomis peltastes*), white crappie (*Pomoxis annularis*), black crappie (*Pomoxis nigromaculatus*), and redear sunfish (*Lepomis microlophus*) were also examined and found to have less parasite diversity than the former two host species. From the three sites sampled, the overall parasite community had more species abundance in the non-urban collection locality than in the two urban sites. Our results indicated that there were significant differences in parasite communities among the three sites, all of which are within 10 km of each other.

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Parasite Mediated Competition between Flying Squirrels (*Glaucomys*)

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Southern flying squirrels (*Glaucomys volans*) are rapidly expanding their range north due to warming temperatures. This range expansion has been implicated in a concurrent reduction in northern flying squirrel (*Glaucomys sabrinus*) range. The reduction in range has also led to conservation concern for northern flying squirrels throughout much of the Great Lakes Region prompting research interest in flying squirrel interactions. The nematode *Strongyloides robustus* is an intestinal parasite infecting most sciurid species. Previous research suggests that *S. robustus* severely worsens northern flying squirrel health while minimally affecting southern flying squirrels. Thus, interactions between the two species are influenced at least in part through parasite mediated competition. *G. volans* have traditionally been believed to be the primary reservoir for *S. robustus* in areas of sympatry, with colonization exposing resident *G. sabrinus* populations to the parasite and resultant reductions in *G. sabrinus* populations. Recent research has suggested that sciurid species other than *G. volans* may be serving as reservoirs for the parasite which would require a major rethinking of our understanding of the interactions between the two species. Squirrels will be tested for *S. robustus* via a modified McMaster fecal float technique, from which parasite eggs are identifiable in the feces. Squirrel trapping will take place through 2023 and 2024. This research aims to determine the prevalence of *S. robustus* among sciurid species in areas where both northern and southern flying squirrels are present in northern Minnesota, and where only northern flying squirrels are present in northern Minnesota. This project is still a work in progress, but only one positive sample of 94 suggests that *S. robustus* and parasite mediated competition do not play a significant role in this system. The conclusions we draw will further inform the ecology of how northern and southern flying squirrels indirectly interact between themselves, other sciurids, and a parasite of concern.

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Flea fauna of Mongolian pikas, jerboas, and zokors

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Despite their medical and veterinary importance, knowledge of the distribution and host associations of most flea species is poor. Documenting the biogeography and ecology of fleas can improve our understanding of host dispersal, disease transmission dynamics, and environmental change. Recent site-intensive surveys of small mammals and their parasites in Mongolia provide an opportunity to investigate patterns of flea diversity across ecosystems and mammalian hosts. Mongolia lies at a major zone of ecological transition from the xeric Gobi Desert in the south through steppe biomes to boreal forests in the north. Documenting the complexity of mammal/flea interactions and their global distributions supports future systematic, ecological, and epidemiological studies, and provides an important baseline for understanding the impact of ecological disruption in a region that is already experiencing severe effects of ongoing climate change. To explore how flea community composition changes with geography and host, we are characterizing diversity and abundance of flea species

associated with Mongolian pikas, jerboas, and zokors. These small mammals inhabit desert, steppe, and alpine environments, and include fossorial, semi-fossorial, and non-burrowing species. Some of these species form large colonies, while others are solitary. We extracted DNA from representative flea species in preparation for future genomic sequencing, and cleared their exoskeletons for morphological identification using published keys. Fleas identified from the burrowing pikas *Ochotona dauurica* and *Ochotona pallasi* and the rock-dwelling pika *Ochotona hyperborea* include genera from the families Pulicidae, Hystrichopsyllidae, Leptopsyllidae, and Ceratophyllidae. Five out of twelve genera identified are primarily associated with pikas: *Amphalius*, *Callopsylla*, *Ctenophyllus*, *Ochotonobius*, and *Paramonopsyllus*. The genera *Amphalius* and *Ctenophyllus* were common to all sampled pika species. Other genera (*Amphipsylla, Echidnophaga, Frontopsylla, Paradoxopsyllus, Peromyscopsylla, Rhadinopsylla*) recovered are host-generalists or are primarily associated with rodents occupying similar habitats. *Echidnophaga*, *Paradoxopsyllus*, and *Paramonopsyllus* were only recorded on *O. pallasi* in far western Mongolia. *Ochotona dauurica* and *O. hyperborea* occurred in close proximity near Lake Khuvsgul, and shared more flea genera (*Amphalius, Ctenophyllus, Frontopsylla*, *Rhadinopsylla*) than expected due to their different life histories. Many fleas are specialized for particular habitats, but ecological disruptions can facilitate exchange of fleas between typically isolated host species.

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Bartonella **diversity in small mammals of Montana and Idaho**

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Species of the gram-negative bacterial genus *Bartonella* are intraerythrocytic parasites of many mammal species, including humans, yet little is known about the distribution and diversity of species in this group. This investigation into the prevalence and diversity of *Bartonella* in wild mammal communities serves as a foundation for understanding the ecological epidemiology of these bacterial pathogens. This is particularly important in ecosystems that are particularly sensitive to ecological disruption associated with climate change, such as high-elevation montane environments. This study focuses on the diversity of *Bartonella* species in small mammal communities in the mountains of Montana and Idaho, with the goal of quantifying the prevalence of *Bartonella* species in different mammal species. These data will document patterns of association between mammalian and *Bartonella* diversity, as well as patterns of diversity along elevation gradients. This investigation includes 25 mammal species (including, mice, voles, shrews, squirrels, pikas, and mustelids) from 18 montane locations (9 in Montana and 9 in Idaho). DNA extracted from frozen liver tissues archived in the Northern Museum of Zoology at Northern Michigan University provide template DNA for droplet digital polymerase chain reaction (ddPCR) to detect if *Bartonella* DNA was present within the target mammal species. Primers and probe specific for *Bartonella* species 16S ribosomal RNA gene were used for ddPCR. Droplet digital PCR was used for *Bartonella* detection due to its high sensitivity to the target DNA, even in low concentrations. Samples that test positive for *Bartonella* by ddPCR were investigated further to determine which *Bartonella* species was present. Following *Bartonella* detection with ddPCR, standard PCR reactions to amplify the citrate synthase gene (*gltA*) were performed. Bidirectional sequencing of the *gltA* gene was performed after PCR amplification. This gene was selected due to its high variability across the *Bartonella* genus. Preliminary ddPCR results showed an overall prevalence of *Bartonella* in 37% (n=143) of small mammals sampled from Montana and Idaho, with 14 of 25 mammal species infected. Preliminary sequencing results show the presence of *Bartonella quintana* and *Bartonella vinsonii* in mammals of this region.

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Digitizing a Parasite Collection at a Primarily Undergraduate Institution: Challenges and Opportunities

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Natural history collections are essential to numerous disciplines in biology and digitization increases access and research productivity. However, many smaller, specialized collections housed at universities present challenges for digitization efforts even though they may yield important discoveries. The Stephen J. Taft Parasitology Collection in the University of Wisconsin – Stevens Point Museum of Natural History was established in 2004 when Dr. Stephen (Steve) J. Taft retired after 33 years of teaching and research at UWSP. The mission of the Parasitology Collection is to serve as a foundation for research and training, primarily of undergraduates, in parasitology and related scientific and applied disciplines. The collection was largely acquired and amassed through the work of Steve Taft and the numerous students he mentored over the decades of his tenure at UWSP. In 2020 faculty-mentored student curation and digitization of the arthropod components of the collection began with the support of the National Science Foundation through the collaborative Terrestrial Parasite Tracker Project. Challenges associated with digitization at a primarily undergraduate institution include limited time from faculty mentors due to high teaching responsibilities, lack of other museum management staff, and the increased training demands and turnover of undergraduate students. Solutions to these challenges include clear and consistent training materials, standardized workflows, peer-mentorship, meticulous recordkeeping, and quality control processes. Helping students develop research ideas related to museum collections helps maintain engagement in the sometimes tedious and repetitive tasks, allows creativity, and builds their confidence through giving presentations. Over four years 21 students contributed to the digitization of 9280 specimens including high resolution slide images and label data to the Ecdysis data portal for use by data aggregators like iDigBio and ultimately the larger community of scientists and educators. This digitization effort has ignited an interest in the UWSP arthropod museum specimens and supported the development of new research efforts aimed at comparing modern collections with historical records including ectoparasites of Great-Horned Owls, Barred Owls, Northern Saw-Whet Owls and other birds of prey. These new projects demonstrate the opportunities for new research projects resulting from access to digital collections.

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Parasites of fishes from the Gladstone and Isaac-Sekulmun Lakes in the Yukon: Implications for a proposed water diversion and hydroelectric power project

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The inter-basin transfer of parasites can have serious consequences for the biota and overall environmental health of a system. A proposed, large-scale water diversion between two separate chains of lakes in the Yukon to generate hydroelectric power prompted a study of the fish parasites of these lakes (2010-2012). In total, 212 fish were necropsied, including 76 Lake Trout (*Salvelinus namaycush*), 55 Artic Grayling (*Thymallus arcticus*), 85 Round Whitefish (*Prosopium cylindraceum*) and 1 Lake Whitefish (*Coregonus clupeaformis*). Morphological and molecular data (DNA sequences of the rRNA gene array and COI gene) were used to identify 24 species of eukaryotic parasites: Microsporidia (1 sp.), Myxosporea (3 spp.), Monogenea (3 spp.), Trematoda (4 spp.), Cestoda (5 spp.), Nematoda (3 spp.), Acanthocephala (1 sp.), Crustacea (3 spp.) and Hirudinea (1 sp.). Of these, the microsporidian and one *Myxobolus* sp. from grayling appear to be new to science. Several parasite species showed host preferences, namely separate species of *Tetraonchus* (Monogenea) in Round Whitefish and Arctic

Grayling, *Eubothrium salvelini* (Cestoda) and *Cystidicola farionis* (Nematoda) in Lake Trout, and *Phyllodistomum* (*umblae*) (Trematoda), *Henneguya* sp. (Myxosporea), and the microsporidian in Round Whitefish. Parasite species recorded in this study corroborate the histological data. With one exception (the piscicolid leech), all species of adult parasites found in Gladstone Lake were also found in the receiving waters of Isaac Lakes and Sekulmun Lake. The close geographical proximity of the two chains of lakes, the similarity of their fish fauna, and their possible connection in the geological past explain our findings.

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Beneath the Surface: Understanding Parasitic Fauna in Dam-Altered Ecosystems

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Dams, primarily constructed for water reservoirs and flood control, significantly alter ecosystems, causing habitat modification, species extirpations, and changes in community composition. This study aims to understand how parasite communities respond to such disturbances using the Mountains to Metro Biodiversity Collection at Kennesaw State University. By examining fluid-preserved fish specimens collected above and below the Allatoona Dam in Georgia (Coosa River Drainage), we investigate the impacts of large-scale disturbances on freshwater fish parasitic fauna, particularly focusing on helminths. Fish specimens are examined internally and externally for parasites, preserving museum integrity by carefully cataloging and removing parasites while minimizing specimen damage. To date, necropsies have been conducted on 148 fish for the project, with an expected estimated total of around 400. It is anticipated that similar to the loss of biodiversity observed among fishes above dams in reservoirs, parasites will undergo a significant decrease in biodiversity. Additionally, it is predicted that generalist parasites will outcompete those with high host specificity, as evidenced by shifts in abundance and prevalence; this may be a result of changes in water temperature due to the thermal effluent of Allatoona Lake, a warm reservoir, or it may be a result of the homogenization of the fish community. Furthermore, increased chemical and nutrient pollution resulting from human activities may exacerbate these effects. The breakdown of seasonal cycles induced by reservoir damming may impede parasites' ability to complete their life cycles by hindering their access to the next host. Through analyses including nonmetric multidimensional scaling (NMDS) and traditional parasite ecology measurements, we aim to elucidate the effects of reservoir damming on parasitic organisms. Understanding the response of parasitic communities to reservoir damming contributes to a more complete comprehension of ecosystem dynamics. By sharing preliminary data, this poster aims to begin shedding light on the often overlooked role of parasitic organisms in anthropogenic disturbances and inform conservation and management efforts aimed at preserving ecosystem health.

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Ectoparasite communities in Barred owls (*Strix varia*) using new and archived museum collections

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The Barred owl (*Strix varia*), originally from eastern North America, has been expanding its territory to the west, including states along the West Coast. Their habitat currently overlaps with the endangered species, the Northern Spotted Owl (*Strix occidentalis caurina*). Barred owls are larger and more aggressive than Northern Spotted Owls, thus making them a superior competitor for food and habitat resources. This expansion further allows hybridization to occur between these two species. This new interaction could lead to exposure to diseases caused by accidental ectoparasites that can be transmitted between the two species. The first objective of this research is to evaluate Barred Owl ectoparasite community richness in its original range in Wisconsin, USA, over time using new and archived museum specimens. To analyze the historical community, I will use samples of slide-mounted museum specimens from 1980-2000 in the Stephen J. Taft Parasitology Collection in the UWSP Museum of Natural History. I will compare these specimens to current slide mounts collected 2022-present and re-evaluate the Barred owl ectoparasite community richness. These results will illustrate the changes in the parasite community of Barred owls due to habitat and population changes in the original range. The second objective will be to assess which ectoparasites found on the Barred owls have been reported from Northern Spotted owls and others that could negatively affect the Northern Spotted Owl's fitness, using museum databases and a systematic literature search. Our museum records include 53 Barred owl specimens, of which 21 have been identified as *Kurodaia magna* and 2 from the genus *Strigiphilus*. Using the 23 specimens identified so far, we can conclude that at least 39.62% of the specimens collected between 1980-2000 are *Kurodaia magna*. Between 2022-present, we collected 37 lice and 6 mites from six barred owls from Wisconsin. We will use morphological traits to identify these specimens and the 30 remaining museum specimens. This research will help identify the potential threat of ectoparasites to the Northern Spotted Owl population, which is decreasing due to numerous threats.

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Parasites of the American Woodcock (*Scolopax minor*) from Wisconsin, USA

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The American Woodcock (*Scolopax minor*) is a migratory shorebird with a unique ecological niche. In contrast to typical shorebirds in the family, it inhabits young forests and fields in eastern North America rather than shoreline habitats. Their diet consists primarily of earthworms and other invertebrates. These associations introduce ample opportunity for interactions between parasites and woodcock. Our study examined primarily hunter-donated woodcock specimens collected in Wisconsin for the documentation of parasites. Feathers were examined for ectoparasites, and internal organs were separated and examined for endoparasites, which were collected and identified based on morphology using 10-60x magnification. Upon collection, parasites were preserved in 80% ethanol. Additionally, liver samples collected and preserved in 95% ethanol were analyzed using molecular techniques to identify haemosporidian infections. Museum specimens from the Stephen J. Taft Animal Parasitological Collection provided a historical baseline and basis of comparisons of parasite identification. Of the 137 archival museum specimens, 57% were nematodes *Tetrameres* spp., 15% other nematode species, 10% digenean species, and 18% phthirapteran species. Of the 28 woodcock specimens necropsied thus far, nematode infections were found in 15 (53%), cestode infections in 21 (75%), digenean infections in 16 (57%), mites in 1 (3%), and lice in 1 (3%). Nematode *Syngamus trachea* was found in one specimen; these nematodes could pose potential harm to the host. *Tetrameres* were present in 5 woodcocks. We screened 18 liver samples for haemosporidian parasites, all of which returned negative results. Molecular and morphological analyses to identify parasites to lower taxonomic levels are underway. Studying these parasites will improve our understanding of host-parasite dynamics in American Woodcock, while also advancing our knowledge of the bird's life history and ecology. These results ultimately will inform conservation efforts for this species which has experienced population declines in recent decades.

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Proposed Project: Can a native toad, *Anaxyrus terrestris, become infected* with the nonnative nematode, *Rhabdias pseudosphaerocephala*

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Invasive species pose a great risk to ecosystems worldwide. When invasive species enter a new ecosystem, they also bring their natural parasites. Non-native parasites pose a great risk to biodiversity due to their ability to spill over into native species. Currently, cane toads, *Rhinella marina*, are one of the most common invasive species in Florida ecosystems. Their lung tissue contains a highly prevalent nematode known as *Rhabdias pseudosphaerocephala* that may spillover into native anurans. The native southern toad, *Anaxyrus terrestris,* is a close taxonomical relative of cane toads and has a very similar nematode parasite that lives in the lung tissue. This proposed study is to determine the ability of the nematode *R. pseudosphaerocephala* to spread into native southern toads by purposely infecting southern toads with the infectious larvae of the nematode found in cane toads. To remove any confounding variables, parasite-free southern toads will be obtained by capturing wild tadpoles and raising them through metamorphosis in the laboratory. *R. pseudosphaerocephala* will be obtained by capturing wild adult cane toads and maintaining them in the laboratory. Feces containing the eggs of the parasite will be placed in a petri dish lined with filter paper to facilitate the development of eggs into L1 larvae, which then sexually reproduce to generate infectious L3 larvae. Metamorphosized southern toads will be exposed to infectious larvae by placing the toad into a small enclosure a with filter paper substrate that is lined in toad feces containing the infectious larvae and left for an 18-hour period to facilitate transmission. Simultaneously, a control group will be set up with the same variables but not receiving the parasite. Assuming transmission has occurred in the exposed group, southern toads will be monitored and cared for 90 days. During this period, southern toads will be weighed three times a week to track any possible weight loss due to infection. After 90 days, southern toads will be euthanized and examined for the presence of adult nematodes in the body to confirm if colonization occurred.

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A novel form of eggstrings for European nematomorphs

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Horsehair worms (Nematomorpha) lay enormous quantities of eggs in the form of large strings. Three different types of such strings were reported up to date and they seem to be genus-specific. In the genus *Paragordius*, strings are very long and thin. In the genus *Gordius*, strings are thicker and tend to fragment into smaller pieces. In the genus *Chordodes*, strings are attached to vegetation in meandering lines. This has been observed for African and North American species. We observed for the first time egg-deposition in *Gordionus violaceus* in Germany. While males are found free-floating in small forest streams, females tend to hide under flat stones at the ground of streams, where they lay eggstrings in meandering lines at the underside of these stones. The similarity of eggstrings in the genera *Chordodes* and *Gordionus* is striking. However, so far this is the only record from the genus *Gordionus* and as stones are not present in all potential habitats, further records of eggstrings in this genus are very interesting.

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Collecting *Nectonema*, the marine Nematomorpha

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In preparation of a genomic project on Nematomorpha, we collected *Nectonema munidae* in Bergen, Norway. We show how *N. munidae* is dissected from its host, the crab Munida tenuimana. Fertilized eggs developed spines upon contact with seawater and first cleavage steps were documented within the eggs.

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Remarks on Indian Gordiids (Nematomorpha)

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The knowledge on Indian Nematomorpha has increased significantly over the past years, although it still may be regarded as fragmentary. Twenty-two species of the phylum Nematomorpha from India have been described so far. The presence of this group was almost negligible for about a century (1912 to 2004), except of only a single report of an undetermined gordiid worm from Darjeeling (in 1986). Most Indian species were reported by Camerano until 1912 and only 13 species of gordiids from 5 genera (*Gordius*, *Chordodes*, *Parachordodes*, *Paragordius*, *Gordionus*) were known from India then. Description of many species was insufficient or just preliminary until then. Since 2004, 9 further species were newly described or newly recorded. 6 of these newly reported species belong to the genus *Chordodes*, and 2 to the genus *Acutogordius.* Also an interesting Gondwana genus, *Beatogordius nagalandis* has been added in India 2017. Incidentally, all new recent additions to gordian species belong to NorthEast India, and other regions of India are still remain under-sampled. Therefore, considering the size of the country and its ecogeographical and climatic diversity, the current number of gordian species from India is only a fragment of the existing biodiversity. With the size of India and its diversity of habitats, a significant diversity is expected, with many endemic species.

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Seroprevalence and molecular characterization of Bovine Anaplasmosis in Bangladesh

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Bovine anaplasmosis is a tick-borne disease in the tropical and subtropical regions in the world. The aims of the study were to reveal the seroprevalence, and characterize and identify the risk factors associated with *Anaplasma* infections in cattle. A total of 384 blood samples were collected randomly in Bangladesh and screened by microscopic examination, cELISA tests and multiplex PCR for the presence of *Anaplasma* species in cattle. The positive amplicons were further confirmed by amplification of the *Anaplasma* species-specific primers like the *groEL, 16s rRNA*, and *MSP2* and sequenced commercially. The sequences were reanalyzed through BLAST in the NCBI database. The microscopic examination reveals 26.82% overall prevalence where the seroprevalence was estimated 43.2% using cELISA and 11.0% prevalence was detected by multiplex PCR from 100 blood samples. The cELISA tests reveals high seroprevalence of *Anaplasma* species in dairy cattle (46.39%) followed by calves (41.08%) and significantly ($p<0.05$) high seroprevalence was found more in cattle (aged>1 year) (52.76%); crossbreed cattle (46.05%) than indigenous cattle breed (34.04%), respectively. The study reveals that calves are also susceptible to *Anaplasma* infections. The multivariable logistic regression analysis identified the age (>1 year), sex (female) and breed (crossbreed) of cattle as potential risk factors. The BLAST analysis reveals that *A. marginale, A. centrale*, *A. bovis*, and *Candidatus A. cinensis* shows 92.47%, 99.78%, 95.51% and 99.29% similarities with the nucleotide sequences of USA and China respectively. Out of the nine sequences results, *A. marginale* was 44.5% prevalent followed by

A. bovis (33.3%), *A. centrale* (11.1%) and *Candidatus A. cinensis* (11.1%). The phylogenetic tree of *16S rRNA* sequences reveals that *A. centrale* clustered together with the *A. centrale* isolate of Philippines. The sequences of *Candidatus A. cinensis* clustered together with *A. platys* C22 and *Candidatus A. cinensis* isolate of Malawi and Bangladesh, whereas *groEL* sequences of *A. bovis* were clustered together with the *A. bovis* isolates of China. The phylogenetic tree reveals that the *A. marginale* of *MSP2* sequences clustered with *A. marginale* of Saint Maries and Florida strain of the USA. Bovine anaplasmosis is very endemic in the study areas and the high number of *Anaplasma* infections suggests a growing concern for animal health and their production.

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Transcriptional profiles of sporulation in species of *Eimeria* infecting chickens

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Eimeria parasites cause enteric disease in livestock and have a devastating impact on poultry production. Oocysts of these coccidian parasites undergo maturation (sporulation) before becoming infectious. To better understand this process, we assessed transcription in maturing oocysts of three important parasite species: *Eimeria acervulina*, *E. maxima*, and *E. tenella.* We investigated predicted gene function and determined which of these parasite's genes also occur in the related human parasite *Cyclospora cayetanensis*. We collected sporulating oocysts every 4-12 hours for 24-48 hours, subjecting each biological replicate to RNA-Sequencing (yielded at least ~0.6 billion paired-end reads, over >84% of which mapped to their reference genome). We designated as "constitutively expressed" those genes always comprising >1,000 transcripts per million (TPM). *E. acervulina* and *E. maxima* shared 41/55 (75%) of such genes; these include included cation-transporting ATPases and housekeeping genes. By contrast, only ten constitutively-expressed genes (of 8,581 annotated genes) were identified in *E. tenella*. Eight of these had homologs in *E. acervulina* and *E. maxima,* in which they were also constitutively-expressed. These genes encode actin proteins, an oocyst wall protein, a profilin, and a rhomboid domain-containing protein, among others. We then identified differentially expressed genes (DEGs) (>1,000 TPM in immature or mature oocysts and undergoing >1.5 or <-1.5 $\log_2$ fold change between these stages). Fewer than 30 genes characterized immature oocysts of *E. acervulina* and *E. maxima* strains, more than 100 characterized immature oocysts of *E. tenella*. Only three such genes (annotated as subtilisin or hypothetical proteins) had homologs in all three species. Over 50 DEGs characterized mature oocysts of *E. acervulina*; fewer such genes were identified for *E. maxima* but more (95) were identified in *E. tenella*. The three species shared ten transcripts characteristic of mature oocysts, including genes encoding micronemes, myosin, profilin, and a group of hypothetical protein-encoding genes. These data illuminate processes influencing sporulation in *Eimeria* and related genera, such as *Cyclospora*, and identify biological processes which may differentiate them. Identifying common developmental pathways may help devise tools to diagnose viability, benefiting animal and public health.

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Fasciola-Infiltrins as a Target for Vaccination

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Fasciola and the common liver fluke are two names used for the same parasite. The infection caused by this parasite is called fascioliasis. *Fasciola hepatica* has been recognised since the pharaonic era and there is evidence of it in mummies; this was the first fluke discovered from the Trematoda class.

Fascioliasis has been reported to be a zoonosis. *F. hepatica* infection has been reported to affect around 17 million people, with a further 180 million at risk in more than seventy countries around the world. It has been reported that *F. hepatica* can infect humans via the consumption of aquatic plants, which often grow near water contaminated with Fasciola from infected animal's faeces. The infection of cattle (approximately 300 million) and sheep (approximately 250 million) results in an annual loss of US$3 billion in affected countries, with the most affected being the owners within the animal husbandry industry. Infiltrins, or pathogen-secreted host nucleus infiltrating proteins, are potential targets for the development of more efficient vaccines against helminthic parasites. Here, we analysed *Fasciola hepatica* protein sequences through use different bioinformatics tools to predict proteins that would show dual CSS/NLS signals. Secondly, the potential vaccine candidate was expressed and purified in our laboratory by using HEK293-EBNA cells and the expression vector pCEP4. Nuclear localisation of fluorescence confirmed the existence of a single, monopartite C-terminal NLS in one of the *F. hepatica* proteins. The predicted FhGST-si candidate 'LKKRAKT' NLS motif. Use of an anti-his tag antibody shows that wild-type recombinant of the potential vaccine candidate, added exogenously to HEK293 cells, fully translocated to the nucleus, whereas the Alanine NLS mutant remained in the cytoplasm. Overall, the existence of an infiltrin in *F. hepatica* suggests that infiltrins may represent a more general regulatory principle operating in parasite.

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Utilizing Natural Products to Inhibit Leishmania Growth and Amastigote Formation by Modifying Essential Host Pathways in Human Macrophages

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Leishmaniasis is a vector-borne tropical disease transmitted by sandflies, caused by a protozoan parasite that spans over 20 different *Leishmania* species, and is contracted by 1 million people each year, killing 50,000 people annually. Visceral leishmaniasis (VL) is the deadliest of three forms of leishmaniasis that causes damage to the liver and spleen and when left untreated results in death. VL is caused by *Leishmania donovani* (LD) and *Leishmania infantum* (LI). Infection occurs after the transmission of extracellular promastigotes from the insect vectors (*Phlebotomus or Lutzomyia* sandflies) and these parasites infect phagocytic cells, such as macrophages and neutrophils, and survive as intracellular amastigotes within the phagolysosome of cells. Current treatments for VL infections include liposomal amphotericin B, miltefosine, paromomycin and pentavalent antimonials. Although these compounds can be effective, they have drawbacks including toxicity to patients and increased resistance. Therefore, new small molecule treatments for this disease are urgently needed. Our recent work indicates that natural products including plant metabolites can be quite effective antileishmanials. Sulforaphane (SFN), is an isothiocyanate agent derived from cruciferous vegetables such as broccoli. SFN has the potential to modify or enhance many host-derived anti-parasitic pathways. SFN activates the NRF2 pathway by modifying redox-sensitive sulfhydryl groups on its inhibitor, KEAP1, to up-regulate the expression of cytoprotective and antioxidant genes. The NRF2 pathway is also involved in inducing autophagy in human cells. We recently published the effects of SFN on extracellular promastigote viability and intracellular amastigote infections in human macrophages that indicate SFN significantly decreases promastigote viability and is highly effective against both LD and LI. The inhibitory effects of SFN against the intracellular amastigote form of LD was assessed post-infection and demonstrated a significant reduction in the number of amastigotes. Recent data in our lab indicates that SFN induces high levels of autophagy with a significant increase in LC3 staining in treated cells and increased levels of P62, which is an adaptor protein involved in selective autophagy. These data support the hypothesis that SFN modifies host autophagy to limit intracellular amastigote replication and provides a new, translational therapy with limited toxicity to complement current treatments against VL.

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Molecular investigation of Tick-Borne Pathogens in Feral Swine (*Sus scrofa*) from seven states in USA

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Feral swine (*Sus scrofa*) have been proposed as part of the lifecycles of multiple tick-borne pathogens (TBP). This is a result of the increase in feral swine populations and increased tick populations and geographic ranges. Few surveys exist of the TBP relevant to human and veterinary health of feral swine in the US. The aim of this study is to investigate the prevalence of 10 TBP: *Borrelia spp., Cytauxzoon felis*, *Hepatozoon spp.*, *Babesia spp.*, *Theileria spp., Mycoplasma spp., Trypanosoma cruzi, Toxoplasma gondii, Yersinia pestis, and Rickettsia spp*. A total of 90 feral swine blood samples were acquired from 29 counties in 7 different states by the USDA Wildlife Services as part of feral swine control. Blood samples were screened via polymerase chain reaction for the detection of the aforementioned TBP DNA. The result indicated the presence of two *Mycoplasma spp*.in 81% of the samples. The 16S rRNA gene sequence analysis (170-193 bp) indicates a close relationship to the *M. suis/M. parvuum* cluster. The presence of *Babesia spp.* was not detected in any of the feral swine samples. The presence of *Babesia* in feral swine hasn't been reported in the United States. These initial results are the first on feral swine in the 7 states. Our investigation will continue to screen feral swine samples for additional TBP and investigate feral swine as a potential reservoir of TBP.

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Quantifying parasite-induced severity of Black Spot Syndrome in *Acanthurus tractus* on reef sites in Bonaire

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Trematode parasites in the genus *Scaphanocephalus* (Opisthorchiidae)have been linked to Black Spot Syndrome (BSS) in Caribbean fish communities. In light-colored fish such as the ocean surgeonfish (*Acanthurus tractus*), metacercariae present as relatively large, melanized black spots or lesions in the epidermis tissue and fins. Recent studies have shown a positive correlation between the number of spots observed externally and the number of encysted metacercariae present, offering a non-invasive opportunity to assess infections in situ. In the current study, research was conducted using video transects taken by scuba divers at 22 reef sites along the western and eastern coasts of Bonaire in the southern Caribbean. Each *A. tractus* individual was recorded and any black spots present on the body were counted by two independent reviewers. Spots that did not resemble the traditional shape or opacity of BSS were not counted. Using individual fish infection, we calculated the average infection prevalence and average spot count for each site and depth. Across the 125 video transects, BSS severity varied from an average of <1 spot per fish to upwards of 15 spots per fish. A higher average lesion count was detected at 2 m (as opposed to 5 m), and BSS was also greatest near the urban center of Kralendijk compared to the less populated sites along the eastern coastline, which also experiences higher currents and wave exposure. Two sites along this eastern coastline that had BSS averages of 0 spots per fish included East Lighthouse and Baby Beach. While sites on the western coastline, such as ChaChaCha, had BSS average values of 11 spots per fish at 2 m depth and 9 spots per fish at 5 m depth. Concordance between independent reviewers in assessing BSS severity was high, averaging 0.85. Patterns of infection are compared to abiotic and biotic characteristics of the dive sites and to previous surveys conducted of the same locations in 2017 to evaluate potential drivers and any temporal changes. Finally, we evaluate associations between BSS and spatial patterns of the coral disease Stony Coral Tissue Loss Disease, which recently arrived in Bonaire.

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Melittobia digitata Parasitoids and Host Preference – Examining how Absence of Preferred Hosts affects Host Selection

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Parasites depend on their preferred host for reproductive success, but when the preferred host is not available some species will parasitize alternative hosts. The study aims to examine the host selection behavior of *Melittobia digitata*, commonly referred to as WOWbugs, when in an environment where the preferred host is not available. WOWbugs are a species of parasitoid wasp which is known to use larvae, the Larger Mud Dauber wasp, as its natural host. In the event that Mud Dauber wasp larvae are not available, the Melittobia digitata is known to use larvae of other species as hosts. This study examines the types of “non-preferred” larvae adult *Melittobia digitata* will infect and with what frequency they will successfully parasitize the alternative hosts. The study will examine Blow Fly larvae, Blue Bottle Fly larvae, and Phoenix Worm larvae. Adult female WOW bugs are first bred with adult males, then placed individually into a rearing chamber containing two prepared larvae of interest from the list above. Specimens remain in the rearing chamber for four days minimum, allowing enough time for parasitism of the larvae to take place. A trial is deemed successful if offspring of the original female emerge from the larvae. A Y-chamber will be used to determine which larvae the females prefer as a host. Preliminary trials indicate that females prefer Blow Fly pupae over Blue Bottle Fly pupae. The topic of alternate hosts is particularly relevant in recent times, especially considering the origins of the SARS-CoV-2 infection and how the virus switched from a natural zoonotic host to a natural human host.

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Developing methods to quantify *Schistosoma mansoni* miracidia behavior and responsiveness

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Schistosomiasis impacts over 250 million people worldwide and is caused by a family of parasitic flatworms known as schistosomes. These parasites have a complex life cycle, requiring progression through a snail intermediate host. During their first larval stage, they are called miracidia, and their main objective is to locate and infect their snail intermediate host so they can develop into a human-infective form. Currently, the mechanisms underlying these interactions are unknown. Dysregulation of these sensory abilities could be an efficient disease control measure, but a thorough understanding of miracidia behavior and responsiveness to host cues is a prerequisite for this prevention strategy. The current study sought to develop methods for quantifying *Schistosoma mansoni* miracidia behavior. To accomplish this, miracidia were harvested from infected tissues and input into slide-based and microfluidic-arena-based assay systems. Their behavior was recorded and then quantified with video and data analysis pipelines integrated into the InVision system. Relationships amongst responsiveness to chemical cues at both fixed and varying time points, behavioral variations amongst miracidia originating from different infected tissues, and hatch rate variability amongst those tissues were evaluated. This work lays the foundation for more efficient, high-throughput, and reproducible analyses of miracidia sensory behavior and responsiveness.

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Interrogation of *Entamoeba histolytica* in a Human Intestinal Tissue Microphysiological System

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The infectious burden of protozoan parasites is extremely significant worldwide and some parasitic species, such as *Entamoeba histolytica (E. histolytica)*, are vastly understudied. Despite its high worldwide burden, little is known about the dynamics of *E. histolytica* pathogenesis in the gut. This gap in research knowledge is largely due to the difficulties associated with analyzing host-pathogen interactions and parasite dissemination in applicable models for human disease. In recent years, great strides have been made in the field of microfluidics, allowing for the integration of human organ systems which can facilitate tissue-tissue crosstalk and effectively model these complex microenvironments. We have developed a human microphysiological system of intestinal tissue and a vascular compartment to investigate *E. histolytica* pathogenesis. This system not only models human architecture but integrates the microbiotic environment necessary to accurately represent the microbial interactions that underly *E. histolytica* non-pathogenic and pathogenic infection.

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Assessing Relationships Between Microplastic Abundance and Parasite Load in Freshwater Fish from Raccoon Creek and Pumpkinvine Creek, Georgia, USA

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The integration of microplastics into freshwater and marine environments is an emerging issue impacting ecosystems worldwide. Microplastics are commonly introduced into ecosystems through various sources, including personal care products, synthetic textiles, and the deterioration of large plastics such as water bottles and tires. Previous studies have found that microplastics have a negative impact on fish populations, altering gene expression and physiology, including immunity, reproduction, feeding, and survival. However, the effects that microplastics have on host-parasite interactions in fish are less understood. In the current study, we surveyed about 200 fluid-preserved fish specimens collected from Raccoon Creek and Pumpkinvine Creek in Georgia, USA, between 1999 and 2013. Initially, we screened the external body of the fish for ectoparasites, then removed the stomach and intestines, examining them for ingested microplastics. Subsequently, all of the internal organs, including the stomach and intestines, were examined for macroparasites. We analyzed the parasite load and microplastic occurrence data to search for potential correlations. Preliminary findings will be presented. Discovering if there is a relationship between microplastic ingestion and parasite load is important to understanding the effect that plastic pollution has on fish health. As microplastic pollution intensifies and poses increased threats to marine and aquatic ecosystems, understanding its influence is paramount to achieving effective management and remediation of the issue.

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Parasite prevalence differs in Bluntnose Minnows (*Pimephales notatus*) across central Illinois creeks

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Freshwater environments, particularly creeks, support a rich community of fish and invertebrate hosts. Water quality may have both negative and positive effects on parasite communities. We aimed to investigate how streams of differing water quality impacted parasite communities. This study was conducted to look at the diversity, incidence, and distribution of parasites infecting fish species

(*Pimephales notatus*) in various creeks in Illinois, USA. A total of 305 fish samples from 12 sites were dissected and examined for parasites including external surfaces, gills and internal organs using a stereomicroscope. The parasites were identified to major taxonomic groups (nematode, cestode, trematode). Microscopy and molecular analysis will be conducted in future for more specific identification. Preliminary data showed that most of the fish infected with trematodes and parasite prevalence differed across creeks. The highest prevalence was found in Kikapoo Creek (100%) and the lowest was from an unknown Creek (44%). Parasites were most frequently collected from the stomach (mean intensity 53) and lowest from the gills (mean intensity 0.43). Furthermore, the parasite prevalence is also influenced by water parameters. This demonstrates that biotic and abiotic factors play a complicated role in the assemblages of parasite communities in creek ecosystems. This study sheds light on the ecology of fish parasites in Illinois creek ecosystems, explaining their variety, distribution patterns, and ecological interactions. Understanding the dynamics of fish-parasite interactions is critical for effective fisheries management, conservation efforts, and maintaining ecological integrity in Illinois creek systems.

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Behavioral Changes in Crickets infected with *Paragordius varius*

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Parasites manipulate their host's behavior using both direct and indirect mechanisms to facilitate their transmission. This study examines cricket behavior when infected by the horsehair worm [Nematomorpha] *Paragordius varius.* Adult horsehair worms live in water and deposit eggs that hatch into larvae and encyst in aquatic insects. When these insects move to land, terrestrial insects ingest them, and the cysts develop into juveniles. When the juveniles mature into adults and are ready to emerge, the infected host seeks out water. This study asks if *P. varius* manipulates the behavior of the field cricket (*Gryllus firmus*) and house cricket (*Acheta domesticus*) so that infected crickets are more likely enter water. To test this, we randomly assigned crickets to infected or sham-infected treatments. Crickets were placed in the middle of a sand-filled arena that contained a water-filled Petri dish in one corner and an empty Petri dish in the opposite corner. After acclimating for 5 minutes under an opaque container, the cricket was allowed to move in the arena where its movement was video recorded under red light conditions for 10 minutes. Each cricket was retested four weeks post-infection in the same manner. EthoVision software analyzed differences in behavior between infected and sham-infected crickets in week 1 and week 4. We hypothesize that the infected and sham infected *A. domesticus* and *G. firmus* would show no significant behavioral difference in week 1. However, in week 4 we hypothesize that infected crickets would spend more time in the water zone than sham-infected crickets. Preliminary results show that in week 4, infected *G. firmus* (n = 44) spent an average of 6.19 seconds in the water zone compared to 15.9 seconds for sham-infected *G. firmus* (n = 27). In week 4, infected *A. domesticus* (n = 49) spent an average of 27.31 seconds in the water zone while sham-infected *A. domesticus* (n =23) spent an average of 21.81 seconds. Results are discussed in the context of testing parasite manipulation.

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The Effect of Ploidy on the Formation and Antimicrobial Efficacy of Extracellular Traps in the Eastern Oyster

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DNA-derived extracellular traps (ETs) represent a highly conserved component of the innate immune system, recently discovered and observed in all metazoan life. When encountering pathogens, cellular DNA is decondensed and released, forming net-like meshes that can capture and neutralize viruses, bacteria, fungi and parasites (both intra and extracellular). ETs are critical to early infection responses regulating inflammation cascades, and encapsulation processes. However, improperly controlled ET production can lead to substantial host harm, elevate tissue damage, and prevent healing. Consequently, the greater DNA content observed in triploid oysters may improve the effectiveness of ETs in combating pathogens or make them more vulnerable to stressors. Numerous stimuli were assessed for their efficacy in generating ETs in the eastern oyster and, once identified, were used to compare the capacity of half-sibling triploid and diploid oysters to generate ETs. We evaluated ET formation and efficacy through a combination of fluorescence microscopy and bacterial neutralization assays, along with corresponding DNAse controls. Our findings demonstrated that inducing a puncture wound in oysters significantly increased the rate of ET formation, which was able to neutralize >60% of introduced bacteria. Additionally, ET efficacy was correlated with the proportion of agranulocytes in hemolymph and the production of reactive oxygen species in these cells, suggesting agranulocytes as potent immune effector cells. ImageJ analysis further revealed that triploid oysters produced more ETs compared to their diploid counterparts, displaying a higher frequency of ET-producing cells and larger ETs, although there was no significant disparity in bacterial neutralization between diploid and triploid hemocytes. In summary, this study describes a reliable method for inducing ETs in oysters and identifies crucial host factors influencing ET formation. ETs likely play critical roles in many antiparasitic strategies though this topic has been only lightly touched upon so far with triploid animals posing as a possible useful tool for further exploration of this conserved immune mechanism.

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Feline cytokine response to Cytauxzoon felis infections with different clinical presentations

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Cytauxzoon felis, a parasite primarily transmitted by the lone star tick (*Amblyomma americanum*), has been increasing in prevalence throughout the southeastern and midwestern United States. *Cytauxzoon felis* is the etiological agent for the feline disease cytauxzoonosis, otherwise known as bobcat fever. The intermediate host of *C. felis* is a feline; the primary reservoir hosts are bobcats (Lynx rufus) while the host of main concern in this research is domestic cats (*Felis catus*). The parasite's geographical range has more recently expanded northwards along with the lone star ticks. Cytauxzoonosis' pathogenesis causes decreased appetite, dehydration, lethargy, fever, hemolytic crisis, dyspnea, and icterus, causing a high mortality rate. Previous studies on the feline immune response to cytauxzoonosis note that the CD18 adhesion molecule is upregulated during infections, possibly causing an increase in proinflammatory enzymes as well. One study found that in cases where cytauxzoonosis was fatal, cytokines TNF-α and IL-1β were increased verses non-fatal/asymptomatic cases. In our study, we measured cytokine profiles Fas, IFNγ, IL-1β, IL-2, IL-4, IL-5, IL-8, IL-10, IL-12 and RANTES in domestic cats clinically ill, asymptomatically infected, and known to have recovered from *C. felis*, and compared them to levels found in healthy domestic cats. We did this by collecting blood samples sent to us by vet clinics in the Southern United States, performed PCR to confirm infection, and used a RayBiotech cytokine kit to determine cytokine levels in each of the aforementioned clinical presentations. Our future goals are to help illuminate possible treatment pathways for infected cats in the future, as well as expanding on the minimal research focusing on the feline immune response to *C. felis*.

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Soil-transmitted helminths in the United States: using big data to characterize patients and analyze disease trends

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Soil-transmitted helminths (STH) include species responsible for hookworm disease (HD; *Ancylostoma duodenale* and *Necator americanus*), ascariasis (AD; *Ascaris lumbricoides*), and trichuriasis (TD; *Trichuris trichiura*). In the United States, STH infections have been greatly reduced with anthelmintic medications and improved hygiene and sanitation; however, cases in the United States still regularly occur but there is limited information on the current epidemiology of these infections. We sought to understand the impact of STH infections on human health using big-data analytics of inpatient medical discharge records through time (1998-2020). Data were obtained from the Healthcare Cost and Utilization Project National Inpatient Sample, the nation's largest publicly available inpatient sample. The data represent a complex-weighted sample of records for approximately 35 million hospitalizations annually. We developed a brute-force algorithm to extract International Classification of Diseases codes for STH infections from over 805 million patient discharge records. We investigated patient characteristics and other epidemiological data. We found a mean of 223 (SD = 70.1) cases annually of STH infections, with AD (n = 2599) most common, followed by HD (n = 1809) and TD (n = 716). Mean STH cases were highest ($p < 0.05$) in males for HD (M: 45 vs. F: 33) and TD (M: 18 vs. F: 13), but females were highest for AD (F: 58 vs. M: 51). Age distributions were heavily skewed towards older patients, with half of STH cases 40 years or older, and 9% under the age of four years. Patients were majority white for HD (54%), white or Hispanic for AD (30% each), and Hispanic for TD (35%), with 55% of most recent reported from the south. Mean hospital length of stay averaged 8 days, resulting in an average hospitalization cost of around $75,000. This analysis serves as a case study for using patient record databases as a means of indirect parasitic disease surveillance that may prove to be useful for understanding the sociodemographic characteristics of patients most at risk for these infections and may assist in obtaining population-based temporal estimates of parasite disease burden for underfunded public health surveillance programs.

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Testing the presence of pheromones in horsehair worms (*Paragordius varius*) using an artificial stream

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The horsehair worm, *Paragordius varius*, exhibits the typical lifecycle of a parasite. Its lifecycle starts as an egg in an aquatic environment and hatches into a larva. This larva infects and manipulates an aquatic host which transfers the worm to a terrestrial host. The worm then manipulates the terrestrial host to enter a body of water where the mature worm exits and reproduces. Despite what we know about the life cycle of the horsehair worm, the process by which male and female worms locate each other when it is time to reproduce is unknown. Some theories on how male and female worms find each other consist of communication via pheromones and even by pure chance. To learn more about how male and female horsehair worms locate each other, we will conduct a series of tests using an artificial stream, that simulates small streams found in Nebraska. The first trial will focus on the role stream flow plays in bringing together worms. Male and female hairworms will be placed in a flowing artificial stream, and their encounter time measured. Encounter time will also be measured for male and female worms in the artificial stream when the water is not flowing. A second trial will test for the presence of pheromones by measuring the encounter time among three pairings of worms: male-male, female-

female, and male-female. The final trial will compare the time for male-female worms to encounter each other with the time it takes for two pieces of string to encounter each other. All trials will be conducted with 20 different worm pairings and last a maximum of ten minutes. We hypothesize that if horsehair worms use pheromones to find each other, then male worms will encounter females more quickly in flowing water than in stagnant water, more quickly than male-male pairings and more quickly than two pieces of strings. If horsehair worms do use pheromones, then this information could be used to develop drugs that disrupt mating in closely related parasitic nematodes.

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Investigative Study into Rearing *Chordodes morgani* in a Lab

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Nematomorpha species *Chordodes morgani* is found throughout Nebraska with wood roaches (*Parcoblatta* spp.) as its definitive terrestrial host and mayflies as its aquatic paratenic host. Experiments have been performed to test the effects of hairworms on their host but failed due to a lack of confirmed infections. To investigate this issue, an experiment was designed in which two groups of wood roaches were infected: one with infected fresh mayfly tissue and the other with infected frozen snail tissue. The presence of cysts in each tissue was confirmed before exposure. Over the three to four months it takes for the worms to mature, the roaches were kept with a wet cotton ball for moisture and food pellets for protein intake. Roaches were housed individually, with a partner, or in groups of three based on the relative density of cysts found in the tissue to which they were exposed. Before infection, each roach's weight, length, and width were measured. A total of 133 roaches were exposed: 45 to mayfly tissue and 88 to snail tissue. To date, 100 roaches have died or been dissected and only three roaches were confirmed to be infected: two roaches in which worms emerged (five worms and two worms, respectively) and one dissected roach with numerous juvenile worms. All emergences came from roaches exposed to fresh mayflies. Results suggest that cysts of *C. morgani*, unlike *Paragordius varius*, are not viable after storage in -80°C conditions. A future study will further investigate the viability of *C. morgani* cysts in a -80°C freezer and if snail tissue is a viable method to infect wood roaches compared to the confirmed way of using mayflies.

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Relating hog management practices to nematode infection on organic and pastured pig farms

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Gastrointestinal nematode parasites are an under-studied yet pervasive issue in non-conventional swine production that can result in economic losses for farmers. The purpose of this study was to measure swine parasite infections on organic and pastured pig farms to explore the relationship between parasite infection and farm management practices. Fecal samples were collected seasonally from 9 pig farms in Pennsylvania and the number of *Ascaris suum*, *Trichuris suis*, *Oesophagostomum* spp. eggs per gram of feces (EPG) was measured for each sample. Management data was collected from each farm, including use of natural anthelmintics and biosecurity measures, and manure management practices. EPG was highest for *A. suum* across all farms, followed by *Oesophagostomum* spp. and *T. suis*. EPG for all three nematodes varied with season, as well as with hog age and sex. There were no differences in the EPG for all three nematodes between farms using biosecurity measures and those that were not. Similarly, there were no differences in EPG for all three nematodes between farms with different manure management practices. EPG for all three nematodes was higher in farms that did not use natural anthelmintics.

Results indicate that the use of natural anthelmintics such as apple cider vinegar and diatomaceous earth is related to low EPG for some nematode parasites. Understanding swine parasite management practices to mitigate the costs of infection is very important for organic and pasture hog farmers, and our results suggest that use of natural anthelmintics may be related to low infection as measured by the eggs shed into the environment.

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A miniaturized sand-zero valent iron (ZVI) filter effectively traps the coccidian parasites *C. cayetanensis* and *Eimeria*: Implications for risk mitigation in irrigation water

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Cyclospora cayetanensis is a coccidian parasite that is transmitted by the fecal-oral route and causes intestinal infections in humans. *C. cayetanensis* outbreaks are often linked to produce that has been contaminated with human fecal material, possibly through irrigation water. Surface and groundwater are at risk of contamination by enteric parasites in areas lacking sufficient sanitation infrastructure, and these areas are often rural locations dominated by agriculture. Filtration of irrigation water with inexpensive sand-zero valent iron (ZVI) has been suggested as a tool to mitigate the risk of crop contamination by *Cyclospora* as well as other pathogenic organisms. We evaluated the use of a sand-ZVI gravity filter to remove *C. cayetanensis* and *Eimeria*, a surrogate organism from water. *Eimeria* is closely related to *C. cayetanensis* and we have used this surrogate model previously. We developed a small-scale model filter that mirrored the performance of larger systems while requiring fewer resources. The reduced size of these minifilters allowed us to perform a wide range of tests to study the effect of filter composition, parasite species, oocyst size, and parasite condition (age, fixation, bleach treatment) on filter performance. Oocysts adhered strongly to iron particles in the filters, and experiments using bleached and unbleached oocysts showed that this adhesion was greater when the outer oocyst wall was intact. We showed that *Eimeria* spp. are effective surrogates for *C. cayetanensis* regarding filtration; sand filters removed *C. cayetanensis* and *E. acervulina* at similar rates (55% and 47%) and sand-ZVI filters removed even more *C. cayetanensis* than *E. acervulina* (97% and 89%). We found that under all conditions, sand-ZVI gravity filters were highly effective in removing both *Eimeria* and *C. cayetanensis* from water, allowing 10-fold fewer oocysts from passing through than did sand filters without ZVI. These inexpensive filters have value as a potential tool for reducing the load of coccidia in irrigation water.

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Prevalence and distribution of zoonotic pathogens and parasites in raccoons (*Procyon lotor*) of northern Illinois

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Raccoons (*Procyon lotor*) are mesopredators that are distributed across the entire continental United States. The growing abundance of raccoons has led to heightened concern regarding disease transfer between raccoons, other wildlife, and humans. Zoonoses are attributed to approximately 60% of known human infectious diseases worldwide and 75% of emerging infectious diseases. Urbanization is a major cause of this increase in human infection with zoonotic disease as land development increases the

probability of human interaction with wildlife. Omnivorous animals such as raccoons thrive in urban settings due to the abundance of anthropogenic resources such as food and shelter, placing them in close proximity to humans. In the USA raccoons are known to carry roughly sixteen zoonotic pathogens, but efforts to quantify their prevalence and distribution along a rural-urban gradient are scarce. The goals of this study are to assess the prevalence and distribution of zoonotic pathogens and parasites, namely *Rickettsia rickettsii*, *Borrelia burgdorferi*, *Babesia microti*-like, *Anaplasma phagocytophilum*, *Ehrlichia chaffeensis*, *Trypanosoma cruzi*, and *Baylisascaris procyonis*, in raccoons at 5 study areas in northern Illinois that represent a rural-urban gradient, explore patterns of coinfection, and evaluate the influence of raccoon density on the prevalence of pathogens and parasites. Raccoons will be captured and removed from sites by collaborators during April-June 2022-2024. One hundred raccoons will be necropsied each year (n=300 total) and tissue samples and intestinal tracts will be collected to assess pathogen and parasite prevalence. To quantify pathogen prevalence, DNA will be extracted from tissue samples and resulting DNA will be quantified via qPCR using pathogen-specific primers and probes. To quantify parasite prevalence, intestinal tracts will be examined, and helminths will be separated and sorted for species identification. Out of 128 raccoons examined for parasite presence from 2022-2023, 98% contained parasites and 25% contained *B. procyonis* (raccoon roundworm). The results of this study will allow an assessment of public health risk that raccoons pose to humans and will help inform wildlife management decisions to integrate the health and wellbeing of wildlife and humans.

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The phylogenetic placement of lecanicephalideans parasitizing the cowtail stingray genus *Pastinachus*

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Members of the cowtail stingray genus *Pastinachus* are host to a great diversity of tapeworms across six orders, including the Lecanicephalidea. Due to the recognition of four new species of *Pastinachus* in the last 20 years and taxonomic issues with many previously described lecanicephalidean species reported from *Pastinachus*, the true diversity of lecanicephalideans parasitizing *Pastinachus* was poorly understood. In a survey of lecanicephalideans parasitizing *Pastinachus* based on morphological data we identified a total of 18 species in the polypocephalid genera *Flapocephalus*, *Anteropora*, and *Polypocephalus*, 17 of which appear new to science, as well as a species representing a putative new genus. Specifically, we identified five species of *Flapocephalus* (including the type species *Flapocephalus trygonis*); four species of *Anteropora*; nine tentacle-bearing species preliminarily identified as members of *Polypocephalus*; and one species belonging to the putative new genus. The goal of this study was to examine the phylogenetic placement and interrelationships of lecanicephalideans parasitizing *Pastinachus* globally. Representatives of 11 of the 19 species documented morphologically, as well as representatives of collectively at least six additional species in *Polypocephalus* and *Flapocephalus* recognized with sequence data only, were included in a phylogenetic analysis of 350 lecanicephalideans based on targeted gene capture data for 402 protein-coding loci. Based on these data, members of *Flapocephalus* formed a clade sister to all other polypocephalids, which, in combination with proglottid anatomy, suggests the potential status of *Flapocephalus* as a distinct family. The single species of *Anteropora* included in the analysis grouped among its congeners with the putative new genus as its sister. The tentacle-bearing species parasitizing *Pastinachus* placed in two of 11 distinct clades comprising the non-monophlyetic genus *Polypocephalus*. In fact, one of these clades, *Polypocephalus* Clade II, is comprised solely of tentacle-bearing species parasitizing *Pastinachus.* Supplementary 28S data for three additional species of the 19 species identified morphologically, included an additional species of *Flapocephalus*, and support a 12th clade of *Polypocephalus* sister to *Anteropora* and the

putative new genus. It appears that lecanicephalidean genera parasitizing a single genus, as is the case with *Flapocephalus*, is becoming the rule rather than the exception within the Lecanicephalidea.

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Finding Dory (*Neoechinorhynchus doryphorus*): Van Cleave and Bangham's mystery worm rediscovered in the Florida Everglades

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A survey of intestinal parasites of freshwater fish was conducted across the eastern portion of the Everglades, Florida from May to August 2023. Seven hundred and fifteen fish from 31 species were investigated from 13 sites across three water bodies designated as storm water treatment areas or water conservation areas as well as Lake Okeechobee and surrounding water bodies. Fish were collected through various sampling means and their intestines were examined for parasites in conjunction with a collaborator in Florida. A new species of *Neoechinorhynchus* was documented from *Micropterus* sp.. While the species strikingly resembles the well-known and widely reported centrarchid neoechinorhynchid *Neoechinorhynchus cylindratus* it differs in 2 key features. The proboscis of the new species is markedly wider than that of *N. cylindratus.* The most distinctive feature of the new species is, however, egg morphology. The eggs of the new species resemble those of many other species of *Neoechinorhynchus* in their possession of polar prolongations of the fertilization membrane but differ in that ends of the prolongations expand into a crown-like structure, a unique feature in the diverse genus. A second species of *Neoechinorhynchus* was also documented from *Micropterus* sp., the poorly-known previously described *Neoechinorhynchus doryphorus* that was described by Van Cleave and Bangham in 1949 based on poor material and not observed since. We provide additional morphological data on *N. doryphorus*. The 3rd acanthocephalan encountered is a species of *Neoechinorhynchus* from the Mayan cichlid, *Mayaheros uropthalmus*, which constitutes a new locality record.

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New insights into reptilian coccidan infections from two species of invasive geckos, the Mediterranean house gecko, *Hemidactylus turcicus* and the tropical house gecko, *H. mabouia* from the new world

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The genus *Eimeria* reported from all vertebrate classes was defined by tetrasporocystic, dizoic exogenous oocysts. However, recent molecular and developmental studies on eimeriid coccidia support multiple linages of coccidia with tetrasporocystic, dizoic oocysts. Two of these lineages, *Choleoeimeria* and *Acroeimeria*, found in reptiles contain a suture on the sporocyst and epicytoplasmic development in the gall bladder or intestine, respectively; and are considered phylogenetically distant from the genus *Eimeria* which infect birds and mammals and contain stiedal bodies on the sporocyst. However, there are several species of *Eimeria*-like coccidia from reptiles, which excyst via the suture in the sporocyst wall but contain intracytoplasmic development. These taxa do not fit the definition of *Eimeria* nor the definition of the genera *Choleoeimeria* or *Acroeimeria*. However, no sequence data is available for these *Eimeria*-like coccidia and it is not clear if they represent a currently undescribed *Eimeria*-like genus or should be included in the *Choleoeimeria* or *Acroeimeria*. To address this issue, we examined the coccidia of two species of introduced house geckos (*Hemidactylus turcicus* and *H. mabouia*) from North America, and documented their oocyst morphology, endogenous development and obtained partial 18s rRNA sequences. Our phylogenetic analyses of all available 18s rRNA sequences of *Eimeria*-like coccidia species from New and Old-World lizard species indicated 3 clades, that differed in site of infection (gall bladder or intestine) and development (epicytoplasmic or intracytoplasmic). Our analysis strongly suggests that a third *Eimeria*-like genus of coccidia infects lizard hosts. This work indicates that oocyst morphology is not useful in differentiating between these genera and obtaining oocyst morphology, endogenous development, and sequence data in future *Eimeria*-like species descriptions will be critical in our understanding of their taxonomical position and phylogenetic relationships. Additionally, our partial sequence of the 18s rRNA gene for *Isospora hemidactylus* obtained during this study and resulting phylogenetic analysis supports previous phylogenetic studies that *Isospora* species from lizards are not a monophyletic group and suggests a paraphyletic origin of *Isospora* species infecting lizard hosts.

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Morphological and molecular characterization of *Hepatozoon* (Apicomplexa: Adeleorina) species infecting frogs and snakes across the central and eastern United States

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The genus *Hepatozoon* includes vector-borne, intracellular blood parasites that infect a wide range of vertebrate hosts including frogs and snakes. Species identification of bloodstream forms is difficult because there are few morphological characters to distinguish species and gamonts of genetically different isolates can be morphologically indistinguishable. This is the case for two *Hepatozoon* species that infect North American anurans, *H. catesbianae* and *H. clamatae*. Traditionally these species have been differentiated based on whether they fragment the host erythrocyte nucleus, however recent genetic studies suggest this character does not correlate with specific genotypes. In this study, we characterized *Hepatozoon* spp. infecting frogs and snakes from the east central United States and compared their effects on the host erythrocytes with genotype. We found individuals of 3 ranid species (*Rana catesbeiana*, *R. clamitans*, and *R. sphenocephala*) were infected with 3 *Hepatozoon* spp. Of those, only individuals of *R. clamitans* were infected with *Hepatozoon* spp. that fragmented the host erythrocyte nuclei; and as previously reported, specific *Hepatozoon* genotypes did not correlate with the presence or absence of fragmentation. However, we found no definitive evidence that *H. catesbianae* fragments the host cell nuclei when infecting *R. catesbeiana.* Importantly, and because of mixed infections being common in frogs and the lack of *Hepatozoon* sequence data from all frog host species we suggest discounting this character as a method of species delineation may be premature. We also report and characterize *Hepatozoon* cf. *sipedon* from 3 snake species. Infected erythrocytes in all 3 snake species displayed variation in the extent of cytoplasm clearing. Sequences from these 3 snakes were identical at *ITS-1* and *18S* rRNA (*COIII* was only sequenced from one isolate). In our *18S* rRNA phylogeny, *Hepatozoon* spp. infecting frogs were in a single clade, whereas *Hepatozoon* spp. infecting snakes were found in multiple clades with *Hepatozoon* spp. that infect other hosts including lizards, small mammals, and frogs. This study adds to a growing number of studies that indicate snakes are capturing *Hepatozoon* spp. from their prey, and we discuss the implications of these host captures for life cycle evolution of *Hepatozoon* spp. infecting snakes.

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On the Identity of a Problematic Set of *Neoechinorhynchus* specimens from buffalo (Catostomidae) from Illinois

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There are three species of *Neoechinorhynchus* (Acanthocephela) that were originally described from Buffalo fishes (Catostomidae: *Ictiobus* Rafinesque, 1820): *Neoechinorhynchus australis* Van Cleave, 1931, Neoe*chinorhynchus distractus* Van Cleave, 1949, and *Neoechinorhynchus strigosus* Van Cleave, 1949. During surveys conducted in 2017 and 2019 we were unsuccessful in obtaining these species from their type hosts in their type localities in Mississippi and Tennessee. In 2023 however, we were successful in obtaining a currently unidentified species of *Neoechinorhynchus* from Smallmouth Buffalo (*Ictiobus bubalus*; Rafinesque, 1818) from the Emiquon Nature Preserve near the Illinois River at Havana, Illinois. Our specimens most closely resemble *N. strigosus*. However, there are numerous morphological discrepancies between the type series of *Neoechinorhynchus strigosus* and our specimens from the 2023 survey. Most strikingly, Van Cleave reported in 1949 that mature females of *Neoechinorhynchus strigosus* have a trunk length ranging from 9 to 14.1 mm, with males ranging from 3.5 to 5.5 mm. Our 2023 female specimens, on the other hand, range from 17.5 to a startling 27 mm, almost double that of the type series of *N. strigosus*. Males follow this trend, with our only sample measuring 9 mm in length. There are also discrepancies with the egg length and width, but this metric can be affected by different fixation approaches and is currently under analysis. Additionally, we are investigating the conspecificity of the two sets of specimens used by Van Cleave in the original description of *N. strigosus* given that one set came from *Ictiobus* sp. from Tennessee whereas the other set came from *Catostomus commersonii* Lacépède, 1803 from Wisconsin. We plan on continuing our morphological comparisons, with the eventual goal of determining if *N. strigosus* requires a redescription or if we have discovered an entirely novel species of *Neoechinorhynchus*.

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Chemosensory behaviors of larval *Aedes triseriatus* mosquitos

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Mosquitos are predominant invertebrate vectors of parasitic diseases, including several prevalent public health concerns such as malaria, Zika, and lymphatic filariasis. Much of disease control research focuses on adult mosquitoes, which act as intermediate hosts, transmitting microscopic viruses and endoparasites via blood meals between vertebrate hosts. However, developing effective disease control may require greater focus on reducing survivability in other vulnerable stages of the vector's life cycle, including the larval stages. We investigated the behaviors of early-stage *Aedes triseriatus* larvae, a species native to Wisconsin and the local vector for La Cross virus, in response to primary exposure to chemical attractants. Using high-resolution cameras and 3D printed behavioral arenas, we have recorded larval physical activity. Furthermore, tracking larval movement during recordings was done using a custom analytical pipeline to collect quantitative data. As behavioral data is collected, larval navigation patterns in response to chemosensory cues may be used to inform vector control research such as responses to larvicidal chemicals.

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Investigating *Bartonella* Prevalence of Intermountain West Fleas

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Fleas are obligate parasites that can transmit blood-borne pathogens in wildlife populations, including small mammal communities in the northern Intermountain West. Some fleas have been documented to prefer specific hosts, whereas other fleas are associated with a broader range of hosts. These alternative patterns of host preference, along with the distribution of flea species across elevation gradients, may have implications for the distribution of vector-transmitted pathogens, such as *Bartonella* species, which are blood-borne, gram-negative bacteria. An understanding of these mammal/flea/*Bartonella* relationships may provide insight into potential pathogen spill-over as climate change continues to disrupt ecological communities. I am studying communities of fleas from 25 small mammal species collected from 18 montane localities in Idaho and Montana to better understand the relationship between pathogen diversity, flea diversity, and host specificity. I am extracting whole genomic DNA from 100 fleas, while retaining intact exoskeletons for species identification. To determine the prevalence of *Bartonella* among the fleas, I am using digital droplet PCR, an ultra-sensitive method for detecting low copy number DNA molecules in a sample. To assess the identity of *Bartonella* in positive samples, I will perform standard PCR to amplify and sequence the gltA gene. Sequences will be compared to known reference samples from GenBank using BLAST searches. The collected data will then undergo phylogenetic analysis with maximum likelihood methods. Preliminary results suggest that *Bartonella* is prevalent in diverse flea species parasitizing several small mammal species. My data show that *Bartonella grahamii*, *B. vinsonii*, *B. volans*, and an unidentified genetic lineage that may represent a new *Bartonella* species, are represented within these flea communities. These results suggest that small mammals may be routinely exposed to *Bartonella*, which presumably increases risk of disease in mammalian communities that experience stress associated with ecological disruption.

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Another new genus and species of fish blood fluke (Digenea: Aporocotylidae) from a common fish (white mullet, *Mugil curema*) in Gulf of Mexico, including pathology and phylogenetic analysis

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Striped mullet (*Mugil cephalus* Linnaeus, 1758) and white mullet (*Mugil curema* Valenciennes, 1836) (Mugilformes: Mugilidae) each represent economically and culturally important fisheries in the Gulf of Mexico, especially in Alabama, Mississippi, and Louisiana. Despite many gill and intestinal parasites being reported from these fishes, no record of a fish blood fluke (Digenea: Aporocotylidae Odhner, 1912 *sensu* Warren & Bullard, 2023) infection in the Gulf of Mexico or northwestern Atlantic Ocean mullet exists. Three blood fluke species infect mullets: *Cardicola mugilis* Yamaguti, 1970 and *Plethorchis acanthus* Martin, 1975 infect striped mullet off Hawaii (Central Pacific Ocean) and in the Brisbane River (Australia), respectively, and *Cardicola brasiliensis* Knoff & Amato, 1992 infects Lebranche mullet, *Mugil liza* Valenciennes, 1836 from Sepetiba Bay (Brazil, Southwestern Atlantic Ocean). White mullet were cast netted and examined for blood fluke infections. Live blood flukes were fixed in 10% formalin for morphology and preserved in 95% ethanol for DNA extraction and subsequent phylogenetic analysis. Tissues were also fixed in 10% formalin for histopathology. The new genus and species differs from *C. mugilis*, *C. brasiliensis*, and *P. acanthus* by having the combination of two testes, post-caecal testes, a uterus with straight ascending and descending portions, and a common genital pore. The *28S* sequence for the new species differed from that of *P. acanthus* by 15% (178 bp). The phylogenetic analysis recovered these sequences as sister and Aporocotylidae as monophyletic. Carditis associated with intense infection by the new species comprised endocardial hyperplasia that markedly thickened the cardiac endothelium. Probable dead or deteriorating eggs in the myocardium were encapsulated by granulomas composed of epithelioid histiocytes. Live eggs infected the afferent artery of gill filaments and were associated with varied hyperplasia of the overlying epithelium and, in high intensity infections, hemorrhaging from the afferent artery.

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Phylogenetics of two "related" monotypic digeneans, *Microcreadium parvum* Simer, 1929 and *Postporus epinephali* (Manter, 1947) Manter, 1949

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Presently, both *Microcreadium parvum* Simer, 1929 and *Postporus epinephali* (Manter, 1947) Manter, 1949 are classified in the Apocreadioidea Skrjabin, 1942. The International Commission on Zoological Nomenclature has recently opined that their Article 35.5 should be suspended for a special case suppressing the Apocreadiidae Skrjabin, 1942 to junior synonym of Megaperidae Manter, 1934. This contentious action reclassified both *M*. *parvum* (Apocreadiinae Skrjabin, 1942) and *P*. *epinephali* (Postporinae Yamaguti, 1958) to the Megaperidae. *Microcreadium parvum* was described from the freshwater drum, *Aplodinotus grunniens* Rafinesque, 1819 from the Tallahatchie River, Mississippi and the species has been reported from drum in inland Ontario and Lake Erie and larval stages are reported in a hydrobiid snail and unionid mussels in Texas. *Postporus epinephali* was described from the red grouper, *Epinephelus morio* (Valenciennes, 1828) and yellowfin grouper, *Mycteroperca venenosa* (Linnaeus, 1758) from Tortugas, Florida. Adults have since been reported from a variety of groupers (Serranidae: Epinephelinae) from Bermuda and the Caribbean Sea. Larval stages are unknown. We collected adults of both species, *M*. *parvum* from the type-host across a large geographical area, and *P*. *epinephali* from the gag grouper, *Mycteroperca microlepis* Goode & Bean, 1879 from the Gulf of Mexico and generated ribosomal DNA sequences from them. We used the partial 28S rDNA gene to attempt to classify them among the Digenea using separate Bayesian Inference analyses. Results were scary: *Microcreadium parvum* is nested deeply among many *Homalometron* spp., suggesting that *Microcreadium* should be synonymized with *Homalometron*. *Postporus epinephali* belongs in the Lepocreadiidae Odhner, 1905, closely related to species of *Lepidapedoides* Yamaguti, 1970. Genetic isolates of *M*. *parvum* from drum collected in Alabama, Mississippi, New York, and Tennessee indicated the species represents a cryptic complex.

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Dulcitransversotrema n. gen. (Digenea: Transversotrematidae) and first morphological and nucleotide-based confirmation of the life cycle of a transversotrematid in North America

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We propose *Dulcitransversotrema* n. gen. for the freshwater transversotrematids *Dulcitransversotrema patialense* (Soparkar, 1924) n. comb. and *Dulcitransversotrema chauhani* (Agrawal and Singh, 1960) n. comb. *Dulcitransversotrema*is unique by the combination of lacking an oral sucker and an extensively coiled uterus as well as having testes that are not deeply lobed and that abut or nearly abut the inner posterolateral margins of the digestive tract (vs. deeply lobed testes that abut the cyclocoel region of the digestive tract only or that do not abut the ceca), a pre-oral genital pore (vs. post-oral genital pore), an ovary that is anterior to and abuts or nearly abuts the sinistral testis (vs. an ovary that is separated from the sinistral testis by some distance or having an ovary that is median and immediately pre-testicular), non-embryonated eggs (vs. embryonated eggs), typically having none or sparse vitelline follicles between the testes and ceca (vs. having a wholly inter-cecal vitellarium comprising a pair of highly compacted clusters of vitelline follicles anterolateral to the testes or having numerous vitelline follicles between the testes and ceca), an oblong, median and primarily inter-testicular vitelline reservoir

(vs. an extensively elongate, pre-testicular vitelline reservoir that arches anteriorly around the sinistral testis), and a subterminal excretory pore opening on the dorsal body surface between the level of the cyclocoel and posterior body end (vs. a terminal excretory pore). We describe the redia and cercaria of *Dulcitransversotrema* cf.*patialense* from the red-rimmed melania, *Melanoides tuberculata* (Müller, 1774) (Cerithioidea: Thiaridae) and their corresponding adult from beneath the scales of the zebrafish, *Danio rerio* (Hamilton, 1822) (Cypriniformes: Danionidae). All hosts were sampled from a spring-fed earthen pond private aquaculture facility near Ruskin, Florida. No transversotrematid life cycle was known from North America previously. The large subunit ribosomal DNA (*28S*) and ribosomal internal transcribed spacer 2 (*ITS2*) sequences of *D.* cf. *patialense* from Florida were most similar to those from *D. patialense* infecting red-rimmed melania from Mayagüez, Puerto Rico and differed by 134 and 69 nucleotides, respectively. Both phylogenetic analyses recovered *Dulcitransversotrema* as monophyletic and sister to a clade comprising *Transversotrema* spp. plus *Crusziella formosa* Cribb, Bray, and Barker, 1992.

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What continental drift? Phylogeny, biogeography and host associations of the Proterodiplostomidae

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Crocodilians are an ancient group of reptiles that evolved at least 225 million years ago and inhabited the supercontinent Pangea prior to its breakup. The geographic range of early crocodilians was fragmented and their descendants were separated from each other by continental drift. Subsequent speciation and extinction events have shaped today's fauna and distribution of crocodilians. The same likely happened to their parasites. Although parasites of crocodilians have been studied insufficiently, published research revealed rich, mostly specific fauna of parasites in these reptiles. One of the most common helminths found in crocodilians are digeneans of the family Proterodiplostomidae Dubois, 1936 (superfamily Diplostomoidea). Members of the Proterodiplostomidae are intestinal parasites of their reptilian definitive hosts in tropical and subtropical environments around the world. Although a few proterodiplostomid taxa are known from turtles and snakes, the overwhelming majority of these digeneans are found in crocodilians. Until very recently, almost nothing was known about interrelationships among proterodiplostomids, which prevented exploring their historical biogeography and evolutionary host associations. Almost no DNA sequence data were available from this fascinating group of digeneans. The situation has changed dramatically in recent years with publication of several phylogenetic studies based on significant amount of novel sequence data, as well as descriptions of several new genera and species from the Americas, Africa and Australia. The system of the family has been revised and new keys for identifications of genera were proposed. We provide an overview of the current state of knowledge in proterodiplostomid phylogenetics, biogeography and host associations with a focus on their evolutionary associations with crocodilians. The results based on specimens from 3 continents and 23 proterodiplostomid genera have demonstrated the monophyly of the group. They also provided evidence that at least some of today's proterodiplostomid lineages are very ancient and likely evolved before the break-up of supercontinents, thus reflecting long co-evolutionary history between these parasites and crocodilians. Phylogenetic analyses also demonstrated several evolutionary host switching events among the Proterodiplostomidae. Molecular data helped to re-assess certain morphological characters used in proterodiplostomid taxonomy, which resulted in numerous systematic changes including descriptions of multiple new genera.

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Under the Surface: Understanding Infection Patterns and Morphology of *Haematoloechus* spp. in California Bullfrogs

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Amphibian populations have declined globally in response to stressors such as land use change, infectious disease, and invasive species, however, the interactive or potentially additive effects of such factors are not well-understood. This study characterized patterns of infection by the multi-host trematodes, *Haematoloechus* spp., which infect the lungs of invasive bullfrogs (*Rana catesbeiana*) in two stock ponds within the California Bay Area. In addition, we clarified confusion regarding species-specific physical attributes of *Haematoloechus* spp. by using DNA sequences and morphological study to develop a local dichotomous key and confirm that, among the ninety-five examined hosts, multiple *Haematoloechus* spp. were present in the California system. We evaluated the individual and interactive effects of host traits on patterns of *Haematoloechus* spp. infection and found that host size positively influenced both *Haematoloechus* spp. prevalence (the parasite's presence or absence) as well as infection load (the number of *Haematoloechus* spp. present) in a frog. By better understanding parasites like *Haematoloechus* spp. which utilize complex life cycles, especially as they interact with other factors changing the ecosystem (invasive species), we will be better equipped to address conservation efforts and recognize other, potentially more complex, systems in the future.

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Molecular phylogeny reveals history of host switching events in the evolution of *Neodiplostomum* (Digenea, Diplostomidae)

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Digeneans of the family Diplostomidae usually parasitize fish- or frog-eating birds and mammals. Some of the most frequently reported diplostomids belong to the broadly distributed genera *Neodiplostomum* and *Fibricola*, the latter often found in ecological and parasite survey studies, most commonly from their amphibian second intermediate host and reptilian paratenic hosts. The two genera are extremely similar morphologically and usually separated based on their definitive hosts (*Neodiplostomum* spp. typically parasitize birds; *Fibricola* spp. are normally found in mammals). Members of the Neotropical genus *Lophosicyadiplostomum*, parasitic in passeriform and cuculiform birds, are morphologically similar to *Neodiplostomum* spp. except for the presence of a muscular apical structure associated with the oral sucker. We collected a diversity of *Neodiplostomum*, *Fibricola*, and *Lophosicyadiplostomum* spp. from North and South America, including the type-species of *Fibricola* and *Lophosicyadiplostomum*. Newly generated sequences of nuclear ribosomal and mitochondrial DNA were used to examine the phylogenetic interrelationships among these genera and other diplostomids, as well as re-evaluate their systematics. Our molecular and morphological analyses prompted the synonymization of *Fibricola* and *Lophosicyadiplostomum* with *Neodiplostomum*. Molecular phylogenies demonstrated at least two independent host-switching events between avian and mammalian hosts in the evolutionary history of *Neodiplostomum*. Our data also indicated that likely a single host-switching event has resulted in a subsequent radiation and diversification of *Neodiplostomum* parasitic in passeriform and cuculiform birds. Some species of this lineage possessed a unique apical structure

developed to varying extents. We also described two new *Neodiplostomum* spp. and re-described two others.

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A plea to develop a convention to share datasets through universal data repositories

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For over 100 years American parasitologists have documented the natural history, evolution, transmission and distribution of parasites. These results were published as checklists, systematic inventories, characterization of infections and parasite communities, among others. Because these summaries are designed by convention among scientists, most are synthetic and exclude metadata, such localities, dates, and environmental conditions for individuals. Availability of metadata would enable the re-utilization of datasets in contemporary analyses which may help us understand changes in parasite transmission and evolution. The metadata is often present in the original fieldnotes and in the last 40 years it has been condensed in spreadsheets. These datasets are accumulated through the professional trajectory of parasitologists and are seldom reused or repurposed. The need to reanalyze data is important because dwindling populations of vertebrates make further sampling impossible or unethical; further, their analysis may help us to identify the causes of decline. Raw data can be a powerful resource for current and future biologists and it may amplify the results presented by their original authors. I advocate for the dissemination of datasets. I suggest that parasitologists: a) make their datasets publicly available through institutional -not private- libraries; b) include data to meet Biodiversity Information Standards; c) link their data to materialistic evidence of the parasites, such as specimens or sequences available for reexamination, and d) provide a citation of the publication featuring the original analysis of the dataset. Users of the universally available datasets must consider crediting the original authors by citing the original source, alternatively, users of the data should consider the role of original authors following the Contributor Roles Taxonomy. Clarity in these practices will motivate scientists to share data as it occurs for DNA sequences and genomes. Further, data dissemination is becoming a requirement from Federal and private granting agencies.

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Tigers with spots: Severe clinostomid outbreak with high morbidity in *Ambystoma tigrinum* from Boulder, Colorado

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One of the most important yet largely unexplored questions is under what conditions the effects of macroparasites on individual hosts translate into population-level consequences. Trematodes in the genus *Clinostomum* form large metacercariae that can sometimes achieve high numbers in their second intermediate hosts, creating the potential for pathology. Here, we characterize a morbidity event in tiger salamanders (*Ambystoma tigrinum*) from a freshwater pond in Boulder, Colorado, linked to clinostomid infection. After using classical necropsy, molecular analysis, and histopathological assessment to record the intensity and pathology associated with infections, we assessed the validity of using non-invasive, image-based methods to quantify infection. Over two years, we collected 42 larval *A. tigrinum* for necropsy and recorded 62 videos. During the high-infection year, 2022, there was a 100% (15/15) infection prevalence of *Clinostomum marginatum*. The following summer, only 11% (3/27) of salamanders were infected with *C. marginatum*. Infections by *C. marginatum* in 2022 had a record-high average intensity (2,406 ± 85 SE; range: 279 to 4,075). Histopathology results indicated that *C. marginatum* was found in nearly all body tissues and organs of the salamanders and was closely associated with hemorrhaging and chronic inflammation around the cysts. All evaluated *A. tigrinum* were co-infected with additional parasite taxa, including the trematodes *Ribeiroia ondatrae* (78.6%) and *Cephalogonimus americanus* (73.8%), as well as the nematode *Chabaudgolvania* sp. (69.0%). Molecular testing for *Ranavirus* and *Batrachochytrium* sp. was negative. Infection loads of *C. marginatum* quantified indirectly via video and were strongly correlated with direct counts via necropsy and were also consistent between independent reviewers, highlighting the utility of this method for non-invasive, non-lethal assessment of parasite infection in these hosts. This extreme clinostomid morbidity event is extremely rare for Colorado, given that this event seemed to have only occurred for a single summer. We will continue monitoring this pond to distinguish whether this event was novel or has the possibility to disturb future salamander populations.

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Parasite prevalence differs in Bluntnose Minnows (*Pimephales notatus*) across central Illinois creeks

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Freshwater environments, particularly creeks, support a rich community of fish and invertebrate hosts. Water quality may have both negative and positive effects on parasite communities. We aimed to investigate how streams of differing water quality impacted parasite communities. This study was conducted to look at the diversity, incidence, and distribution of parasites infecting fish species (*Pimephales notatus*) in various creeks in Illinois, USA. A total of 305 fish samples from 12 sites were dissected and examined for parasites including external surfaces, gills and internal organs using a stereomicroscope. The parasites were identified to major taxonomic groups (nematode, cestode, trematode). Microscopy and molecular analysis will be conducted in future for more specific identification. Preliminary data showed that most of the fish infected with trematodes and parasite prevalence differed across creeks. The highest prevalence was found in Kikapoo Creek (100%) and the lowest was from an unknown Creek (44%). Parasites were most frequently collected from the stomach (mean intensity 53) and lowest from the gills (mean intensity 0.43). Furthermore, the parasite prevalence is also influenced by water parameters. This demonstrates that biotic and abiotic factors play a complicated role in the assemblages of parasite communities in creek ecosystems. This study sheds light on the ecology of fish parasites in Illinois creek ecosystems, explaining their variety, distribution patterns, and ecological interactions. Understanding the dynamics of fish-parasite interactions is critical for effective fisheries management, conservation efforts, and maintaining ecological integrity in Illinois creek systems.

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Effect of coat color on parasitism in eastern gray squirrels

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Phenotypic variation in host color may result in differences in exposure to parasites and the ability to resist established parasites. Although these relationships are increasingly being recognized, they remain poorly understood. Melanism, whereby certain individuals have increased dark pigmentation in their plumage and pelage is of particular interest as melanin-based coloration has been linked to changes in

both behavior and immunity. Eastern gray squirrels (*Sciurus carolinensis*) are good models for exploring the relationship between melanin-based coloration and parasitism as there are typically two color morphs, gray and melanic (black), controlled by a single gene. Though the mechanism is unknown, recent studies on coat color in grey squirrels have shown that urbanization increases the prevalence of melanic squirrels. Increased pressure from parasites in densely populated urban areas may drive selection for the melanic morph. In this study, we quantified differences in ectoparasite loads between grey and melanic morphs to determine if coat color influences susceptibility and resistance to ectoparasites. We captured melanic (n=30) and grey (n=48) squirrels across three sites in Syracuse, NY and combed each for ectoparasites. We found no correlation between parasite load and color morph. This indicates that melanin-based coloration does not confer an ectoparasite-based advantage to urban grey squirrels. This research adds to the growing body of literature exploring the relationship between coloration and parasites.

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Disrupting host sensing of the myxozoan parasite *Ceratonova shasta*

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Myxozoans are spore forming cnidarian endoparasites, responsible for fish diseases including whirling disease, enteronecrosis, and proliferative kidney disease. Like their free-living cnidarian relatives, myxozoans have an organelle called the nematocyst, consisting of a tubule that discharges explosively when triggered by specific stimuli. In free-living cnidarians, nematocysts are used for capturing prey and deterring predators, but in myxozoans they are used for attaching to their hosts to begin infection. The precise factors that trigger myxozoan nematocysts remain largely unknown. We propose that an understanding of the host-sensing mechanism could lead to a method for blocking the parasites and preventing infections. Possible host receptors in myxozoans include a diverse range of carbohydrate moieties, present in different organelles and most importantly on the spore surface. In this study, we are probing different carbohydrate moieties using the Vector Labs Lectin Kit I, containing ConA, DBA, PNA, RCA120, SBA, UEA I and WGA. We're examining actinospores and myxospores of the myxozoan *Ceratonova shasta*, a parasite that causes enteronecrosis and death in both wild and hatchery-reared salmon and trout. We predict that the surface-expressed moieties we identify are part of the host-sensing mechanism. We will then test their role in host sensing by selectively blocking or inhibiting them and observing results in *in vitro* and *in vivo* experiments.

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Could pinworms be mutualists in fiber digestion?

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Pinworms are typically regarded as having minimal impact on their hosts due to a relative lack of pathogenicity. In lizards that rely on plant-based diets, however, heavy pinworm burdens have been suggested to have a positive impact on host digestion. Dense infections could increase digestive efficiency if pinworms digest plant matter in their own guts, selectively feed on host gut microbes, and/or host their own gut microbiomes that enhance plant matter degradation. We investigated the contributions of pinworms to digestion in herbivorous Bonaire whiptail lizards (*Cnemidophorus ruthveni*), which are known to host heavy pinworm infections. If pinworms enhanced digestion in the suggested ways, we hypothesized that pinworms would be isotopically enriched compared to host tissue and digesta due to selective feeding in the host gut and that pinworms would harbor a gut microbiota

that differed in composition from the host gut microbiota. We trapped lizards near Boka Onima on the Caribbean Island of Bonaire to collect lizard tissue, intestinal parasites, and large intestine contents. We identified parasites to species level and assessed total worm burden, determined whether worms were relatively enriched for carbon and nitrogen stable isotopes, and compared the microbiotas of the host large intestine and worm intestine (16S rDNA metabarcoding) to better understand host-parasite relationships in digestion. All lizards hosted the pinworms *Alaeuris rinconensis* and *Ozolaimus megatyphlon* in their large intestine, and two lizards also hosted the cestode *Oochoristica iguanae* in their small intestine. The minimum pinworm burden was >100 worms, and infections frequently included immature and mature specimens of both pinworm species. Upon completion, stable isotope analyses will reveal the trophic relationship of host and parasites, and microbiome profiling of pinworm and host gut tracts will reveal whether worms harbor unique gut microbes or maintain gut microbes in relative abundances that differ from the host. This work will be an important contribution to understanding the feeding ecology of pinworms and elucidating whether they can provide a digestive benefit to herbivorous hosts that do not have a specialized gut fermentation chamber.

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Characterization of an apicomplexan parasite associated with the collapse of the bay scallop (*Argopecten irradians*) population in New York

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Since 2019, bay scallop populations in New York have been suffering large-scale summer mortalities resulting in over 90% reduction in biomass. These events were associated with heavy infections by an undescribed parasite. The objective of this research was to characterize this parasite and elucidate factors that regulate disease dynamics. Microscopic and molecular investigations showed that the parasite is an undescribed member of the *Apicomplexa*, with its closest relative being a member of the newly established class *Marosporida*. The parasite, dubbed Bay Scallop Marosporida (BSM), infects and disrupts multiple scallop tissues including kidney, adductor muscle, gill, and gonad. Both intracellular and extracellular stages of the parasite were identified, including sporozoite stages that display structural characteristics of the *Apicomplexa* (e.g., micronemes). Field surveys since 2020 demonstrated a strong seasonal signature in disease prevalence and intensity, with severe cases increasing as summer progresses before slightly decreasing, as a likely result, at least in part, of the mortality of most heavily infected scallops. Field and laboratory investigations demonstrated the role of prevailing environmental factors and scallop genetic background on disease dynamics and resulting mortalities. For instance, laboratory investigations underline higher mortalities in most severely infected scallops exposed to ecologically-relevant high temperature, while dissolved oxygen alone appears to have less effect on disease dynamics. Interestingly, both laboratory and field studies showed contrasted disease severity among different scallop stocks with up to 4-fold difference in scallop survivorship. Overall, our results suggest that BSM infection plays a major role in the collapse of bay scallop populations in New York, possibly by synergistically interacting with stressful environmental conditions to impair the host and lead to mortality. Current work includes unraveling the ecology of the parasite and sequencing its genome to facilitate in-depth investigations of host-parasite interactions.

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Parasite communities of critically endangered hosts are influenced by different sites of translocation as revealed by metabarcoding

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Translocation is one of the most important tools for conserving endangered animals. However, conservation-driven translocation, aimed at helping the host species, often neglects symbiotic organisms such as parasites and may even lead to parasite losses, extirpation and extinction. Here, we examined nematode and coccidia parasites of translocated populations of the critically endangered woylie (*Bettongia penicillata*) found in four sanctuaries across Australia (Perup Sanctuary, Mt Gibson, Scotia and Yookamurra). Faecal samples of woylies were collected at each location in 2019. We aimed to compare the parasite communities of these four populations via metabarcoding of faecal samples using taxon-specific metabarcoding primers. While each location had a similar component community diversity, we found that woylies in different sanctuaries had distinct parasite infracommunities in terms of prevalence of infection, mean infracommunity diversity, and infracommunity composition. Contrary to our expectations, estimates of parasite prevalence and diversity were not strongly related to host density, geographic diversity of founding host population or time since establishment of the sanctuary. Instead, parasite prevalence and diversity appeared to be influenced more strongly by stochastic environmental factors during and after translocation, so that woylies in different translocation sites had distinctly different parasite communities despite considerable overlap in source populations. Such differences could potentially lead to displacement, extirpation or even extinction of certain rare or endemic parasites if they cannot be maintained in translocation sites, with repercussions for both the health of the host and the wider ecosystem.

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Anthelmintic efficacy of *Trigonella foenum graecum* seed extracts against *Isoparorchis hypselobagri* (Digenea: Trematoda) infecting swim bladder of freshwater cat fish *Wallago attu*

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Helminth infections in aquaculture present significant challenges due to concerns associated with conventional anthelmintic drugs, including environmental impact, host toxicity, and residue contamination. The current study is an attempt to address these challenges by investigating the anthelmintic potential of *Trigonella foenum graecum* (fenugreek) seed extracts against *Isoparorchis hypselobagri*, a trematode parasite infecting freshwater catfish, *Wallago attu*. Crude Fenugreek seed extracts (aqueous, methanol, ethanol and ethyl acetate) were prepared using the soxhlet extraction method. Qualitative and quantitative phytochemical screenings identified alkaloids, flavonoids, phenolics, saponins, tannins, terpenoids, coumarins and glycosides. Anthelmintic activity against *I. hypselobagri* was assessed *in vitro* at 1, 10 and 20 mg/ml concentrations. Positive (albendazole) and negative (RPMI-1640 media) controls were employed and LC50 values were calculated. Methanol extracts demonstrated the highest extraction yield (11.8%) and exhibited higher levels of phenolics, flavonoids, alkaloids, and terpenoids. FTIR analysis confirmed the presence of phenols and alkaloids. *In vitro* anthelmintic assays demonstrated the time and dose- dependent efficacy of fenugreek seed extracts, with the methanol extract showing the highest potency (LC50). Scanning electron microscopy revealed severe tegumental damage, including folding, lesions, erosion and extensive blebbing, underscoring the pronounced anthelmintic potential of methanol extracts. This study emphasizes the substantial anthelmintic potential of *T. foenum graecum* seed extracts, emphasizing the efficacy of the methanol extract against *I. hypselobagri*. The findings underscore the promising alternative these extracts offer for managing parasitic infections in freshwater catfish. Further *in vivo* validation is essential to ascertain the practical application of fenugreek seed extracts as eco-friendly and sustainable phytotherapeutic agents in helminth infection control.

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Ecology of fear: *Drosophila nigrospiracula* prioritizes anti-predator behaviour at the expense of sub-lethal parasitic infection

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Survival in ecosystems that contain multiple natural enemies requires an ability to assess and prioritize relative risks and respond accordingly. This inherently leads to trade-offs, in which a focal species invests more or less time in a given response depending on the associated risk, e.g. foraging less in the presence of a predator. An investigation of trade-offs regarding multiple natural enemies represents the leading edge of non-consumptive effects (NCEs) and the ‘ecology of fear’ research. These theoretical constructs seek to understand the effects of avoiding consumption (in part or whole) by natural enemies and the associated fitness costs of the avoidance strategies. Viewing behavioural NCEs in the context of multiple enemies allows us to assess responses to one threat that may make the focal individual more vulnerable to a secondary threat. Here, we use a novel system to evaluate a two-threat scenario using the fruit fly *Drosophila nigrospiracula*, a parasitic mite, and a predatory spider. Our previous research with this system showed that the fly demonstrated distinct anti-parasite and anti-predator behaviours that were diametrically opposed. Our present hypothesis, that flies prioritise anti-predator behavioural defenses when exposed to both parasite and predator, was tested with two experiments in which flies were (i) directly exposed to parasites with and without odour and visual predator cues and (ii) directly exposed to predators with and without parasite cues. Hypothesis 1 predicts that given the greater fitness cost of predation *vs* parasitism, infection levels would increase when predator cues were present. Hypothesis 2 predicts that the presence or absence of mites would not alter fly predation. We found that when predator cues were present, infection prevalence increased from 17 to 30%, and a four-fold increase in the number of mites attached to flies. In contrast, the presence or absence of parasite cues did not influence the rate of predation by the spider. These results show that *D. nigrospiracula* has the ability to prioritize behavioural defences, trading off predator defense against parasite avoidance. This study has important implications for understanding NCEs in a landscape of fear where multiple threats exist.

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Teeny, Tiny, Tapeworms—They Fit on the Head of a Pin

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A trend has emerged from survey work conducted over the past two decades that challenges aspects of the prevailing concepts of the bounds of tapeworm size. It has long been recognized that the giants of the tapeworm world are members of the orders Cyclophyllidea and Diphyllobothriidea, with some species achieving lengths of over 30 m. But what about the smallest tapeworms? Historically, candidates for this honour—which we would argue should be small enough to fit on the 2 mm head of a pin—consist of cyclophyllideans of birds (e.g., members of the Amabiliidae, Progynotaeniidae, and Dilepididae) and a few mammals (e.g., members of the Hymenolepididae), predominantly in temperate regions. However, our collections from elasmobranchs have yielded 17 species of tiny tapeworms across the orders Diphyllidea, Lecanicephalidea, Onchoproteocephalidea, Rhinebothriidea, and Serendipeidea. Inspection of the literature revealed an additional 6 tiny species in the Onchoproteocephalidea. We see

few commonalities across these 23 species that might account for their tiny size. They parasitize a broad range of hosts (stingrays, guitarfish, or skates). More than half occur in the waters of Southeast Asia; the remainder occur in the Gulf of Mexico, Gulf of California, and parts of the Atlantic Ocean. What do these elasmobranch cestodes have in common with the tiny tapeworms that parasitize birds and mammals in temperate regions? One factor is temperature. Although the tiny cyclophyllideans occur in temperate regions, their bird and mammal hosts are endothermic and have resting body temperatures well above the ambient temperature of their environment. In contrast, almost 80% of the tiny elasmobranch tapeworms occur in tropical waters, in hosts with a body temperature typically reflecting that of their environment. The fact that many animals living in tropical areas tend to have smaller body sizes than those living in temperate areas (i.e., Bergmann's rule) is typically attributed to the fact that smaller bodies help shed heat. Do tapeworms have issues regulating temperature? If so, how might they shed heat? Although the answers are not immediately apparent, understanding the factors that contribute to the small size of these endoparasites can advance our understanding of this phenomenon overall.

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Adaptive evolution of stress response genes in two monogenean parasite lineages aligns with their respective host niche diversity

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Stress responses are crucial for parasite survival and, consequently, contribute to the evolutionary success of these organisms. However, the evolution of the molecular pathways dealing with environmental stressors is poorly understood in parasites. A notable bias in parasite genomic research towards few health-relevant human pathogens results in a lack of DNA and RNA sequence data and limiting knowledge of diversification mechanisms among wildlife parasites. Comparative studies of closely related and functionally similar lineages of species-rich wildlife parasite taxa might provide insight into the factors driving adaptive evolution of stress responses. To address this knowledge gap, we comparatively investigated antioxidant, heat shock, osmoregulatory, and behaviour-related genes (*foraging*) in the two parasitic flatworm lineages with varying species and ecological diversity, *Cichlidogyrus* and *Kapentagyrus* (Platyhelminthes: Monogenea), through whole-genome sequencing of 11 species. We assembled the sequences of 48 stress-related genes using an exon bait capture approach. The study reports the first *foraging* (*for*) gene orthologs in flatworms. We also discovered that species of *Cichlidogyrus* present gene duplications of heat shock (*hsp*) and oxidative stress genes compared to *Kapentagyrus*. This difference might be linked to the high ecological diversity of the host lineage of *Cichlidogyrus* (African cichlid fishes), unlike the ecologically conserved niches of the hosts of species of *Kapentagyrus* (African freshwater sardines). Positively selected sites were detected in genes related to mitochondrial protein import (*hsp*) and behaviour (*for*) in species of *Cichlidogyrus* infecting a host lineage undergoing adaptive radiation (East African cichlids), suggesting an adaptive link to a putative co-radiation of parasites and hosts. Therefore, the study identifies the first potential molecular functions linked to a flatworm radiation. Additionally, the absence of cytochrome P450 and kappa and sigma-class glutathione S-transferases in monogenean flatworms is reported, genes typically considered essential for metazoan life.

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Variation in diplostomid parasite infection patterns and pathology in Bluegill sunfish (*Lepomis macrochirus*) reproductive morphotypes

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Bluegill sunfish (*Lepomis macrochirus*), a common North America sportfish, employ a mating system with multiple male reproductive morphotypes that include nest-building α-males with high reproductive investment and satellite β-males that are nest-parasites with no parental investment. The larger α-males are dominant within the territory and tend to be more successful in their mating success, but despite their reproductive and territorial dominance, α- and β-males occur in equal proportions within most *L. macrochirus* populations. As indicated by previous studies, parasitism differences between male morphotypes may contribute to the unexpected proportions of *L. macrochirus* morphotypes. Diplostomid parasites, particularly *Posthodiplostomum* spp. (white grub) and *Uvulifer ambloplitis* (black spot disease), are amongst the most common parasite species infecting *L. macrochirus*, but their impact on fish health is debated with extensive variability reported on their effects. This study investigated the pathological impact of diplostomid parasites on the health of *L. macrochirus*, if the patterns of parasite infection differed between male morphotypes, and if the health impacts from the parasites differed between host morphotypes. In total, 3,406 *L. macrochirus* were collected from 18 lakes and ponds in northern Virginia. The fish were necropsied to identify and enumerate the diplostomid parasites infecting the hosts. In low abundance, neither diplostomid parasite impacted the body condition of *L. macrochirus*, but high abundances of both *Posthodiplostomum* spp. (> 900 metacercariae) and *U. ambloplitis* (> 40 metacercariae) negatively impacted the health of *L. macrochirus*. There was no difference in the immune response between *L. macrochirus* morphotypes, indicating there may be threshold for parasitism in diplostomid parasites that must be reached before the parasites have a significant impact on fish health. Alpha-males recruited significantly more diplostomid parasites than β-males and had a greater proportion of hosts exceeding the infection thresholds in both parasites. These parasitism differences and impact on host body condition may be contributing to the maintenance of an inferior reproductive morphotype in greater abundance than would be expected based on reproductive output.

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The role of centrachid host diversity on *Posthodiplostomum* spp. infection in Bluegill sunfish (*Lepomis macrochirus*)

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The role of species diversity and disease risk has been spurred by rapid biodiversity losses in the changing landscape. The dilution effect, a leading hypothesis for the importance of biodiversity in disease reduction, suggests that increasing species diversity within a system decreases the risk of disease amongst the organisms inhabiting it. This is particularly true for helminth parasites exhibiting complex life cycles that have a high degree of host specificity. Amongst the most common sportfish inhabiting freshwaters of North America are Bluegill sunfish (*Lepomis macrochirus*), which frequently serve as a second intermediate host for one of the most common centrarchid fish parasites, *Posthodiplostomum* spp. (white grub). While all members of the Centrarchidae are susceptible to this trematode parasite, susceptibility varies between species, indicating varying degrees of parasite dilution that may lessen the parasite burden on *L. macrochirus* hosts. The dilution effect was analyzed in two data sets: (1) a collection of 4,583 centrarchid fish from 18 lakes and ponds in northern Virginia; (2) a

meta-analysis of 13,663 centrarchid fish from 306 lakes and ponds across the continental United States. The prevalence, intensity, and abundance of *Posthodiplostomum* spp. infecting *L. macrochirus* were analyzed to determine if lake surface area, *Lepomis* diversity, and centrachid diversity impacted parasite infection in the focal host. While there were no significant correlations with lake surface area and *Posthodiplostomum* spp. infection in *L. macrochirus*, both *Lepomis* diversity and centrarchid diversity exhibited significant correlations where increases in species diversity resulted in decreased parasite burden on *L. macrochirus* hosts. Additionally, there were varying effects of centrarchid co-occurrence as some fish species had larger diluting impacts than others. This study indicates the importance of species diversity in reducing the parasite burden in one of the more common freshwater sportfish species in the U.S. Additionally, *Posthodiplostomum* spp. only has a significant impact on fish health in high intensities (> 900 metacercariae) and reducing the parasite burden has important consequences for long-term management of *L. macrochirus*.

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Visualization of the host-parasite interface uncovers potential mechanisms of behavior manipulation in *Dicrocoelium*-infected zombie ants

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Understanding the mechanisms that parasites use to manipulate the phenotypes of their hosts remains an elusive goal. One indirect tool is to match particular host manipulations with patterns of parasite site-selection within host tissues. For instance, characterizing the host-parasite interface within regions of the CNS offers a path to understanding parasite-induced manipulations of host behaviours. The results of imaging studies involving the brains of *Toxoplasma*-infected rodents and rabies-virus infected carnivores provide examples of this approach. Larvae of the iconic manipulating trematode, *Dicrocoelium dendriticum*, reside within the brain of their ant intermediate hosts. A single larva in the brain ('brainworm') orchestrates the attachment and detachment of an infected ant's mandibles onto flower petals. This behavior is thought to facilitate larval transmission into the grazing mammals in which the worms mature. We combined traditional histology with CT imaging and 2-photon, confocal, and electron microscopy to characterize the brain/brainworm interface during the period when infected ants affix their mandibles to flowers. Brainworms resided within the ventral- and anterior-most region of the sub-esophageal ganglion (SEG) of the brain, partially enveloped by a thin and flexible bi-layered cyst wall. The outer wall was always in direct contact with host neuropil tissue. The anterior-most parts of some brainworms appeared to extend through the cyst wall to make direct contact with dorsal regions of the host SEG and immediately proximal to regions of the SEG that play a role in host feeding. Our results hint at potential mechanisms, in addition to those that can interfere with the action of the host's mandibular muscles, that likely play a role in the complex manipulation of ant behavior during infection.

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Evolution of a Pathogenic Neogregarine (*Ophryocystis elektroscirrha*) in Milkweed Butterflies

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The pathogenic neogregarine *Ophryocystis elektroscirrha* infects the hypodermal tissues of monarchs (*Danaus plexippus*). Three transmission routes have been proposed for *O. elektroscirrha* and include horizontal transmission, maternal transmission, and sexual transmission. Caterpillars become infected when they ingest oocysts from milkweed leaves or egg cases after hatching. However, the diversity of

these parasites in other milkweed butterflies is not well known. To evaluate this, we examined 64 species of milkweed butterflies from 11 genera and from 6 continents; including all *Danaus* species (monarchs, tigers, and queens) for *Ophryocystis* infections. Based on oocyst morphology and complete ITS rDNA sequences, only 4 species of milkweed butterflies in the genus *Danaus* and from 2 of the 3 subgenera were infected with 3 linages of *Ophryocystis*. Importantly, oocyst morphology and host pathology (cuticle damage) or lack of, was conserved within but distinct among host clades. More interestingly, for maternally and sexually transmitted parasites, the distribution of *Ophryocystis* linages on the *Danaus* phylogeny was peculiar. For example, *Ophryocystis* occurred in 3 of 4 species of queens (subgenus *Anosia*) and showed no cuticular damage, 0 of 4 species of tigers (subgenus *Salatura*; the sister clade to queens) and 1 of 3 species of monarchs (subgenus *Danaus*) with extensive cuticular damage. More importantly, the host phylogeny was incongruent with the *Ophryocystis* phylogeny, suggested a possible evolutionary host switch of *Ophryocystis* from queen butterflies to monarch butterflies. To test this hypothesis, we exposed 2 groups of monarch caterpillars to *O. elektroscirrha* from monarch butterflies, or *Ophryocystis* sp. from queen butterflies. After eclosion the 2 exposed monarch groups along with unexposed controls were placed in cages with honey water and evaluated for infection status, longevity and weight loss. Our results, indicate that monarchs exposed to *O. elektroscirrha*, had a significantly shorter life span and lost significantly more weight than control monarchs; whereas the longevity and weight loss of monarchs exposed to queen *Ophryocystis* species was not significantly different from control monarchs. The implications of our hypothesis are discussed in terms of host switching events for sexually transmitted parasites, and the unique biology of monarch butterflies and their conservation efforts.

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Avian schistosomatids and their life cycles in Southern Cone of South America

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Endemic and migratory waterfowl and aquatic snails were examined for avian schistosomatids (Schistosomatidae) in Chile and Argentina. While studies on mammalian schistosomes are numerous worldwide, there are relatively fewer studies on avian schistosomatids in South America, which causes a zoonotic condition in people. Studies on avian schistosome biology and systematics are emerging in Neotropics, showing a remarkable diversity of endemic schistosomes. A total of 19 waterfowl, including endemic ducks, waterfowl, *Netta peposaca, Spatula versicolor, Cygnus melancoryphus* and a migratory species *Spatula cyanoptera* from different locations in Argentina and Chile were dissected. In addition, 1390 *Physa* cf. *acuta* (Physidae) snails were collected in different water bodies from Central and Southern Chile, and 10 *Physa* sp. were collected in a pond in Argentina. Endemic species of *Chilina* snails were also examined. Two life cycles were revealed using molecular characterizations, that of an unnamed endemic genus Schistosomatidae sp. lineage 2 going through *C. melancoryphus-Chilina dombeiana* in Chile and *Trichobilharzia querquedulae* going through migratory *S. cyanoptera-Physa* sp. in Argentina. Other species found were a species of *Trichobilharzia* sp. from a diving duck *N. peposaca*, *Nasusbilharzia melancorhypha* in *C. melancoryphus*. This study highlights the distribution of schistosomes in birds as both endemic in the birds and *Chilina* spp. snails and highly dispersed species *T. querquedulae* found in its long-distance migratory host, *S. cyanoptera*. Unfortunately, the lack of museum vouchers from previous studies did not allow a comparison of the unidentified lineages found in this study.

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An ecologist's journey in parasite land

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My journey from mediocre undergraduate with zero interest in parasitology to this year's recipient of the ASP Eminent Parasitologist award has been shaped by serendipity, lucky breaks and near misses. It is a story beginning with great mentors and then supported for three decades by a wonderful cast of students, postdocs, research assistants and collaborators. In my address, I will share that journey to date (it is not quite finished yet!), and present some of the research highlights that have brought me here. I will finish with some advice for early-career parasitologists, distilled from my years of experience in the field.

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ASP Meeting History

1925 Kansas City MO
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1942 No meeting
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2014 New Orleans LA
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* With the American Society of Tropical Medicine; since 1952, American Society of Tropical Medicine and Hygiene
ǂ With the American Institute of Biological Sciences
\+ With the Helminthological Society of Washington
§ With the American Microscopical Society
¶ With the International Congress of Parasitology; 1970 (ICOPA-II), 1982 (ICOPA-V), 2002 (ICOPA-X), 2006 (ICOPA-XI)
\# With the Wildlife Disease Associaton
ɸ With the American Association of Veterinary Parasitologists
ƻ With the Society of Protozoologists
Ↄ With the Society of Nematologists
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Ѧ With the Sociedad Mexicana de Parasitología
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¤ With the International Coccidiosis Conference