The 98th Annual Meeting of the American Society of Parasitologists

InterContinental Kansas City at the Plaza

Kansas City, MO July 13–16, 2023

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.

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The American Society of Parasitologists

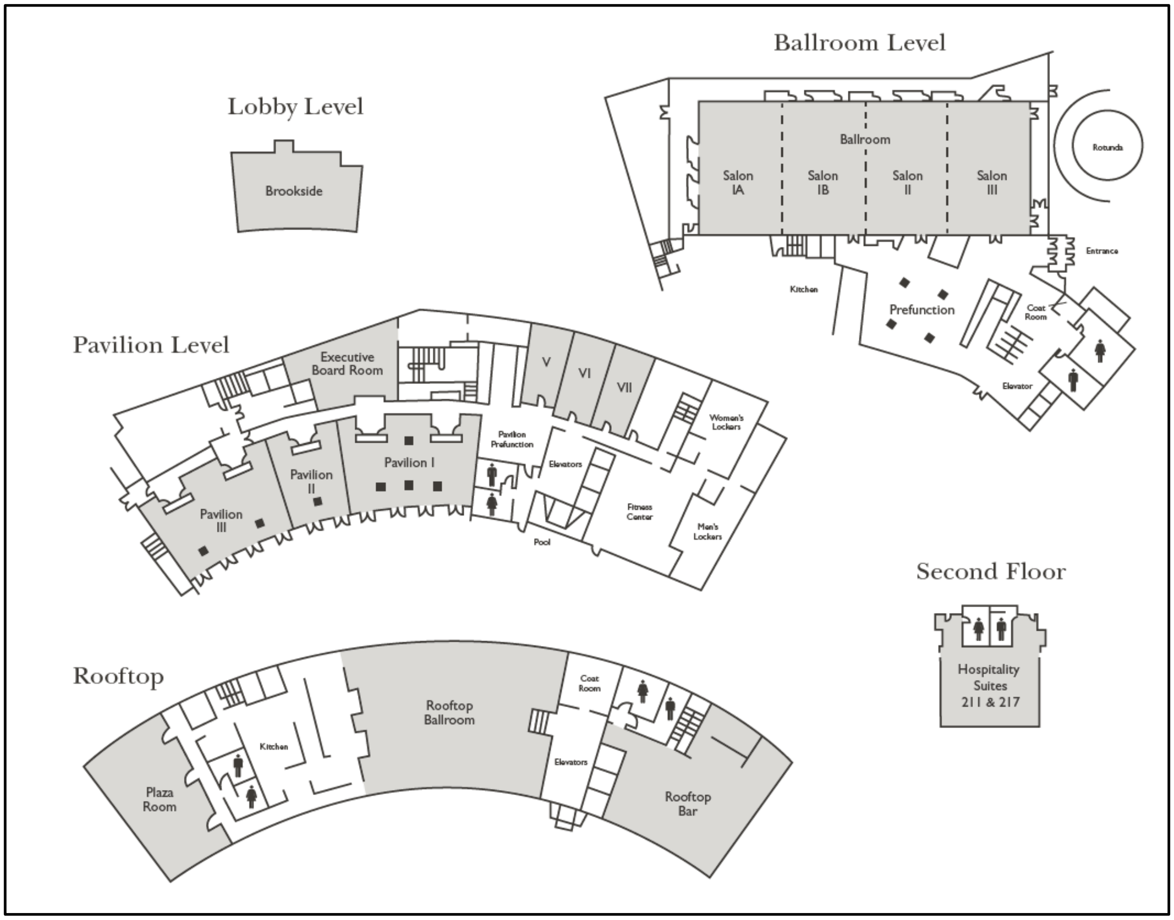
Welcome

We would like to welcome you to the 98th annual meeting of the American Society of Parasitologists (ASP).

The ASP is a diverse group of over 800 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



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
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General Schedule

Day/Times	Activity/Function	Room/Space
Wednesday July 12		
2:00–5:30 pm	Workshops	Plaza Room
Thursday July 13		
7:00 am–5:00 pm	Registration	Ballroom Foyer
8:00 am–Noon	ASP Council Meeting	Pavilion I
1:00–2:30 pm	Life Cycles & Epidemiology	Salon II
1:00–2:30 pm	Ecology & Evolution I	Salon III
2:30–3:00 pm	Coffee Break	Ballroom Foyer
3:00–5:00 pm	Taxonomy, Systematics, & Phylogeny I	Salon II
3:00–5:00 pm	Chemotherapy & Drug Resistance	Salon III
6:00–9:00 pm	Welcome Reception	Rooftop Ballroom & Plaza Room
Friday July 14		
7:00 am–5:00 pm	Registration	Ballroom Foyer
8:20–10:30 am	President's Symposium	Salons II & III
10:30–11:00 am	Coffee Break	Ballroom Foyer
11:00 am-Noon	President's Address	Salons II & III
Noon–1:00 pm	Student Lunch Vortex	Rooftop Bar
1:00–2:30 pm	Taxonomy, Systematics, & Phylogeny II	Salon II
1:00–2:45 pm	Host-Parasite Interactions I	Salon III
1:00–2:00 pm	Vector Biology	Pavilion I
2:30–3:00 pm	Coffee Break	Ballroom Foyer
3:00–5:00 pm	ASP Student Symposium & 3rd Annual Palettes and Parasites Contest	Salon II
6:00–9:00pm	Auction Preview & ASP Student Auction	Rooftop Ballroom & Plaza Room
Saturday July 15		
7:00–11:00 am	Registration	Ballroom Foyer
8:30–10:00 am	Genomics & Molecular Biology I	Salon II
8:30–10:00 am	Ecology & Evolution II	Salon III
10:00–10:30 am	Coffee Break	Ballroom Foyer
10:30 am–Noon	Taxonomy, Systematics & Phylogeny III	Salon II
10:30 am–Noon	Host-Parasite Interactions II	Salon III
Noon–1:00 pm	Editorial Luncheon	Pavilion II
1:15–2:30 pm	Genomics & Molecular Biology II & Immunology	Salon II
1:15–2:30 pm	Ecology & Evolution III	Salon III
2:30–3:00 pm	Coffee Break	Ballroom Foyer
3:30–4:30 pm	Student Business Meeting	Salon III

Wednesday, July 12th

6:00–8:00 pm	Poster Session	Salons IA & B

Sunday July 16

8:00–10:00 am	Insect Physiology & Behavior Symposium	Salon II
10:00–10:30 am	Coffee Break	Ballroom Foyer
10:30 am–Noon	Taxonomy, Systematics & Phylogeny IV	Salon II
10:30 am–Noon	Host-Parasite Interactions III	Salon III
1:00–2:00 pm	H.B. Ward Medal Lecture	Salons II & III
2:15–4:30 pm	ASP Awards & Business Meeting*	Salons II & III

*Awards being presented include the Ashton Cuckler New Investigator Award, Distinguished Service Award, Marc Dresden Student Travel Grants, and Best Student Presentations.

Detailed Schedule

Wednesday, July 12

2:00–5:30 pm Workshops Plaza Room

2:00-3:00 pm **Workshop 1:** Infecting the Media: A workshop on preparing a press release

Featuring C.J. Janovy, Director of Content, KCUR Kansas City; Kay Hawes, Associate Director of News and Media Relations, University of Kansas Medical Center; and Celia Llopis-Jepsen, reporter for the Kansas News Service.

Organizers: Florian Reyda & Sara Weinstein, in conjunction with the ASP Public Relations Committee.

3:30-5:30 pm **Workshop 2:** A practical introduction to generating genomic data using the Oxford Nanopore MinION sequencing platform

Organizers: Kaylee Herzog & Joseph Fauver, University of Nebraska Medical Center.

Thursday, July 13

7:00 am–5:00 pm Registration Ballroom Foyer

8:00 am–Noon ASP Council Meeting Pavilion I

1:00–2:30 pm Life Cycles & Epidemiology Salon II

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

Presiding: **Egie Elisha Enabulele,** Texas Biomedical Research Institute

1:00 (1) **Charles T. Faulkner**, Crystal LeRoy. MICROFILAREMIA IN SHELTERED CANINES INFECTED WITH *DIROFILARIA IMMITIS* IN THE CUMBERLAND GAP REGION OF TENNESSEE, KENTUCKY, AND VIRGINIA.

1:15 (2) **Stephen S. Curran**, John H. Brule, Steven P. Ksepka, Triet N. Truong, Micah B. Warren, Stephen A. Bullard. INVESTIGATING DIGENEAN DIVERSITY IN AN ESTUARINE POND ADJACENT TO THE GULF OF MEXICO ON THE FORT MORGAN PENINSULA, ALABAMA.

1:30 (3) **Elliott A. Zieman**, Barbara S. Carlward, Sofiane Aiche, Alexa Howard, Natalie Mitlyng, Harper Nerbergall, Kacey Newhart, Faith Nuss, Esther Onuselogu, Ryan Pulliam, Cassandra Strehl. *SARCOCYSTIS* IN WILDLIFE FROM THE MIDWESTERN US.

1:45 (4) **Kinfu Yazachew Geremew**. PREVALENCE OF MALARIA AND ITS ASSOCIATED RISK FACTORS IN SELECTED MALARIOUS KEBELES OF GINDEBERET DISTRICT, WESTERN ETHIOPIA: COMMUNITY BASED CROSS-SECTIONAL STUDY.

2:00 (5) † **Tanveer Amhad**, Muhammad Mudasser Nazir, Muhammad Abubakar Siddique. PREVALENCE OF *NEOSPORA CANINUM* ANTIBODIES IN PAKISTANI CAMEL BREEDS (*CAMELUS DROMEDARIUS*) AND ITS EFFECT ON THE ABORTIVE HISTORY OF ANIMALS.

2:15 (6) † **Muhammad Mudasser Nazir**, Ayesha Sarwar, Tanveer Ahmad, Muhammad Mazhar Ayaz. ELISA BASED DETECTION AND PREVALENCE OF IGG ANTIBODIES TO *TOXOPLASMA GONDII* INFECTION IN CLINICALLY HEALTHY PET DOGS FROM SOUTH REGION OF PUNJAB, PAKISTAN.

1:00–2:30 pm Ecology & Evolution I Salon III

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

Presiding: **Whitney C. Preisser,** Kennesaw State University

1:15 (7) † **Sidney C. Mann**, Jillian T. Detwiler. ELUCIDATING THE INFLUENCES OF MICROHABITAT ON GASTROPODS TO PREDICT HOTSPOTS OF BRAINWORM (*PARELAPHOSTRONGYLUS TENUIS*) TRANSMISSION.

1:30 (8) **Daniel C. G. Metz**, Ryan F. Hechinger. THE MOST EXTREME TREMATODE SOLDIER CASTE OCCURS IN A HUMAN-INFECTIOUS, INVASIVE FRESHWATER SPECIES.

1:45 (9) **Colin D. MacLeod**, Christopher D. G. Harley. A WIDESPREAD HOST-PARASITE ASSOCIATION IN THE STRAIT OF GEORGIA PROVIDES A UNIQUE OPPORTUNITY FOR STUDYING THE EFFECTS OF CLIMATE CHANGE ON PARASITISM IN THE MARINE ENVIRONMENT.

2:00 (10) † **Alexandria P. Nelson**, Emily C. Oven, Zachary Gajewski, Trey Jeffers, James Flowers, Jeff Beane, Kenzie Pereira, Skylar R. Hopkins. PARASITE BIODIVERSITY IN SNAKES FROM THE U.S. AND CANADA.

2:15 (11) **Sarah Goodnight,** Michael McCoy, Ellen Titus. WORMS GOT YOUR TONGUE? ORAL PARASITES ALTER MALE ADVERTISEMENT CALLS AND DISRUPT EXPECTED RELATIONSHIPS BETWEEN MALE TRAITS AND FEMALE CHOICE IN GREEN TREE FROGS.

2:30–3:00 pm Coffee Break Ballroom Foyer

3:00–5:00 pm Taxonomy, Systematics, & Phylogeny I Salon II

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

Presiding: **Kaylee Herzog,** University of Nebraska Medical Center

3:00 (12) † **John H. Brule**, Micah B. Warren, Haley R. Dutton, Triet N. Truong, Steven P. Ksepka, Stephen S. Curran, Jacob A. Shurba, Justin D. Krol, Stephen A. Bullard. FIRST TAXONOMICALLY CONFIRMED RECORD OF A GYRODACTYLID, *GYRODACTYLUS CICHLIDARUM* PAPERNA, 1968 (MONOGENOIDEA) INFECTING FARMED NILE TILAPIA, *OREOCHROMIS NILOTICUS* (LINNAEUS, 1758) (CICHLIFORMES: CICHLIDAE) IN THE UNITED STATES.

3:15 (13) † **Jacob A. Shurba**, Matthew L. Horton, Paul D. Johnson, Micah B. Warren, John H. Brule, Triet N. Truong, Stephen S. Curran, Haley R. Dutton, Steven P. Ksepka, Justin D. Krol, Stephen A. Bullard. PARASITOLOGICAL SURVEY OF TWO NEWLY DISCOVERED POPULATIONS OF EXOTIC INVASIVE MYSTERY SNAILS (*CIPANGOPALUDINA*: VIVIPARIDAE: BELLAMYINAE) IN ALABAMA AND ARKANSAS.

3:30 (14) Oluwaremilekun Grace Ajakaye, **Egie Elisha Enabulele**. A NEW LINEAGE OF *ECHINOSTOMA* IN THE "*REVOLUTION* GROUP" SPECIES COMPLEX TRANSMITTED BY *BULINUS* SNAIL IN NIGERIA.

3:45 (15) † **Triet N. Truong**, Nathan V. Whelan, Paul D. Johnson, Michael L. Buntin, Stephen A. Bullard. MORPHOLOGICAL DESCRIPTION, LIFE CYCLE, AND PHYLOGENETICS OF A NEW SPECIES OF *PROTEROMETRA* (DIGENEA: AZYGIIDAE) FROM THE CAHABA RIVER, ALABAMA, U.S.A.

4:00 (16) † **Micah B. Warren**, Haley R. Dutton, Stephen A. Bullard. *NOMASANGUINICOLA DENTATA* (PAPERNA, 1964) WARREN AND BULLARD, 2023 (DIGENEA: SANGUINICOLIDAE POCHE, 1926) INFECTING AFRICAN SHARPTOOTH CATFISH, *CLARIAS GARIEPINUS* (BURCHELL, 1822) TEUGLES, 1982 (SILURIFORMES: CLARIIDAE) FROM THE OKAVANGO RIVER, NAMIBIA: SUPPLEMENTAL DESCRIPTION AND PHYLOGENETIC ANALYSIS.

4:15 (17) † **Zoe S. Von Holten**, Tyler J. Achatz, Alan Fecchio, Stephen E. Greiman, Vasyl V. Tkach. REVEALING DIVERSITY OF THE DIPLOSTOMIDAE (DIGENEA) PARASITIC IN KINGFISHERS.

4:30 (18) † **Steven P. Ksepka**, Triet N. Truong, Stephen A. Bullard. A NEW SPECIES OF *ELLIPSOMYXA* KØIE, 2003 (BIVALVULIDA: CERATOMYXIDAE) INFECTING THE GALL BLADDER OF *PANGASIUS MACRONEMA* BLEEKER, 1850 (SILURIFORMES: PANGASIIDAE) FROM THE MEKONG DELTA, VIETNAM.

4:45 (19) † **Logan S. Elkin**, Jay Bowerman, Michael L. Kent, Anindo Choudhury. MORPHOLOGICAL AND MOLECULAR DESCRIPTION OF A *CREPIDOSTOMUM* SP. (TREMATODA: ALLOCREADIIDAE) AND THE DIVERSITY OF *CREPIDOSTOMUM* FROM THE PACIFIC NORTHWEST.

3:00–5:00 pm Chemotherapy & Drug Resistance Salon III

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

Presiding: **Daniel J. Sprague,** Medical College of Wisconsin
Kayhan Ilbeigi, University of Antwerp

3:00 (20) † **Homa Nath Sharma**, Jonathan Catrett, Ogochea Destine, Audrey Napier, Boakai K. Robertson, Daniel A. Abugri. *IN VITRO* ACTIVITY AND MECHANISMS OF ACTIONS OF METHANOLIC EXTRACT OF *TRAMETES VERSICOLOR* (TURKEY-TAIL) MUSHROOM AGAINST *TOXOPLASMA GONDII.*

3:15 (21) **Daniel J. Sprague**, Sang-Kyu Park, Claudia M. Rohr, Jonathan S. Marchant. THE ANTHELMINTIC ACTIVITY OF PRAZIQUANTEL ANALOGS CORRELATES WITH DIFFERENT STRUCTURE-ACTIVITY RELATIONSHIPS AT $\mathrm{TRPM}_{\mathrm{PZQ}}$ ORTHOLOGS.

3:30 (22) † **Claudia M. Rohr**, Daniel J. Sprague, Sang-Kyu Park, Nicholas J. Malcolm, Jonathan S. Marchant. NATURAL VARIATION IN A PARASITIC FLATWORM ION CHANNEL UNDERPINS DIFFERENTIAL SENSITIVITY OF PARASITES TO PRAZIQUANTEL.

3:45 (23) † **Kayhan Ilbeigi**, Dorien Mabille, An Matheeussen, Rik Hendrickx, Nick Van Reet, Birgit Mertens, Roel Anthonissen, Fabian Hulpia, Louis Maes, Clement Regnault, Phil Whitfield, Marzuq A. Ungogo, Harry P. De Koning, Serge Van Calenbergh, Guy Caljon. DISCOVERY OF A NOVEL THERAPEUTIC CANDIDATE AGAINST ANIMAL TRYPANOSOMIASIS.

4:00 (24) Elise L. McKean, Emilia T. Grill, Damien O'Halloran, **John M. Hawdon**. ALTERED ACTIVATION RESPONSE ASSOCIATED WITH MULTIDRUG RESISTANCE IN THE CANINE HOOKWORM *ANCYLOSTOMA CANINUM*.

4:15 (25) † **Bernard B. A. Efa**, Homa Nath Sharma, Boakai K. Robertson, Audrey Napier, Daniel A. Abugri. CAFFEIC ACID POTENTIATES PYRIMETHAMINE ACTIVITY *IN VITRO.*

4:30 (26) **Nkiru Charity Eberemu**, Nasiru Abullahi. PERSISTENCE OF ANTIGENS OF HISTIDINE RICH PROTEIN II AND III AFTER TREATMENT WITH MALARIA DRUGS.

6:00–9:00 pm	**Welcome Reception**	**Rooftop Ballroom & Plaza Room**

Friday, July 14

7:00 am–5:00 pm	**Registration**	**Ballroom Foyer**
8:20–10:30 am	**ASP President's Symposium**	**Salons II & III**

Presiding: **Tamara J. Cook,** Sam Houston State University

Theme: "Parasitology @ PUI's".

8:20 am	**Tamara J. Cook**: Introduction
8:30 (27)	**Ramon A. Carreno**. PARASITOLOGY TEACHING AND RESEARCH AT SMALL, UNDERGRADUATE INSTITUTIONS: BENEFITS AND CHALLENGES.
9:00 (28)	**Sarah Orlofske**. LEARNING SHOULD BE INFECTIOUS: UNITING TEACHING AND RESEARCH IN PARASITE ECOLOGY AND EVOLUTION.
9:30 (29)	**Florian Reyda**. GETTING AND KEEPING STUDENTS HOOKED ON FISH PARASITE RESEARCH.
10:00 (30)	**Nancy Smith.** RESEARCH FROM DAY ONE: ENGAGING FIRST-YEAR STUDENTS IN RESEARCH @ PUI'S.

10:30–11:00 am	**Coffee break**	**Ballroom Foyer**
11:00 am–Noon	**President's Address**	**Salons II & III**

Presiding: **Joanna Cielocha,** Rockhurst University

11:00 am	**Joanna Cielocha**, Rockhurst University and **Autumn Smith-Herron**, Sam Houston State University: Introduction of Tamara J. Cook.
11:10 am	**Tamara J. Cook**. "BIG SCIENCE AT SMALL INSTITUTIONS: PARASITOLOGY RESEARCH AT PRIMARILY UNDERGRADUATE INSTITUTIONS."

Noon–1:00 pm	**Student Lunch Vortex**	**Rooftop Bar**

Friday, July 14th

1:00–2:30 pm Taxonomy, Systematics, & Phylogeny II Salon II

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

Presiding: **John H. Brule,** Auburn University

1:00 (31) † **Katherine M. Hanselman**, Kirsten Jensen. FLAPS, SPIKES, AND TENTACLES: DIVERSITY OF LECANICEPHALIDEAN TAPEWORMS PARASITIZING COWTAIL RAYS.

1:15 (32) † **Jessica C. Paul**, Janine N. Caira. INTERESTING RESULTS FROM THE “HOTEL CALIFORNIA”: ON THE SPECIES OF *CAULOBOTHRIUM* PARASITIZING STINGRAYS IN THE EASTERN PACIFIC OCEAN.

1:30 (33) † **Valerie R. Mingrone,** Janine N. Caira. MORE THAN JUST A BALL WITH A SKIRT: ADVANCING *BALANOBOTHRIUM* AND ITS RELATIVES TO A NEW ORDER.

1:45 (34) † **Netanya A. Williams**, Janine N. Caira. PERSISTENCE PREVAILS: DO ANY GENERA OF CARCHARHINIFORM SHARKS NOT HOST *ANTHOBOTHRIUM*?

2:00 (35) **Altangerel T. Dursahinhan**, Scott L Gardner. ENDOPARASITE DIVERSITY OF SUBTERRANEAN RODENTS (RODENTIA: MAMMALIA) AND BIOGEOGRAPHICAL HYPOTHESES OF THE GENUS *AROSTRILEPIS* MAS-COMA & TENORA, 1997 (HYMENOLEPIDIDAE: CYCLOPHYLLIDEA), TAPEWORMS FROM THE HOLARCTIC.

2:15 (36) † **René Monzalvo**, F. Agustín Jiménez, Kurt M. Neubig. MOLECULAR CHARACTERIZATION OF *TRICHURIS* SPP. (NEMATODA: TRICHURIDAE) FROM RODENTS AND MARSUPIALS FROM THE NEARCTIC AND NEOTROPICAL REGIONS.

1:00–2:45 pm Host-Parasite Interactions I Salon III

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

Presiding: **Adriana M. Perrucci,** Tarleton State University

1:00 (37) † **Brooke A. McPhail**, Patrick C. Hanington. A LONGITUDINAL SNAIL-TREMATODE SURVEY AT RECLAIMED WETLANDS IN CENTRAL ALBERTA.

1:15 (38) **Matthew Bolek.** AMPHIBIAN MYIASIS IN NORTH AMERICA: AN UPDATE ON HOST USE, DISTRIBUTION AND TAXONOMY OF CALLIPHORIDS THAT INFECT AMPHIBIANS.

1:30 (39) † **Ryan W. Koch**, Giselle C. Ozuna, Matthew G. Bolek. A NOVEL TURTLE DISEASE? HISTOPATHOLOGY OF TURTLES INFECTED WITH ACANTHOCEPHALANS.

1:45 (40) **Sarah Pangburn**, Janus Borner, Susan Perkins. NICHE EXPANSION OF MALARIA PARASITES IN CARIBBEAN LIZARDS.

2:00 (41) **Peter Halvarsson**, Frida Martin, Ylva Hedberg Alm, Eva Tydén. STRONGYLE NEMABIOMES IN HORSES: PREVALENCE, ANTHELMINTIC RESISTANCE, AND FACTORS AFFECTING ABUNDANCE.

2:15 (42) † **Maaz Saleem**, Muhammad Zahid Shah. THE PREVALENCE OF GASTROINTESTINAL PARASITE IN BREEDS OF CATTLE, GOAT AND SHEEP IN NORTHERN PAKISTAN.

2:30 (43) † **Kamila Cajiao-Mora**, Stephen A. Bullard. FIRST PARASITOLOGICAL SURVEY OF BATS (MAMMALIA: CHIROPTERA) FROM THE AMAZON RIVER IN COLOMBIA.

1:00–2:00 pm Vector Biology Pavilion I

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

Presiding: **Jacob A. Shurba,** Auburn University

1:00 (44) † **Wilfred Ouma Otambo**. INFLUENCE OF LANDSCAPE HETEROGENEITY ON ENTOMOLOGICAL AND PARASITOLOGICAL INDICES OF MALARIA IN KISUMU, WESTERN KENYA.

1:15 (45) † **Cole J. Meier**, Julian F. Hillyer. THE TOXICITY OF PHOTOSENSITIVE INSECTICIDES AGAINST MOSQUITOES IS BIDIRECTIONALLY MODULATED BY THE ABUNDANCE OF LARVAL FOOD.

1:30 (46) † **NOTE: This talk was moved to Ecology and Evolution II**

1:45 (47) † **Ashley Aguilar**, Megan Wise De Valdez. MOSQUITO ECOLOGY IN ZOOLOGICAL PARKS: IMPLICATIONS FOR ZOONOSES OF MOSQUITO-BORNE DISEASES.

2:30–3:00 pm Coffee Break Ballroom Foyer

3:00–5:00 pm ASP Student Symposium & 3rd Annual Palettes & Parasites Contest Salon II

Presiding: **Sarah Goodnight**, Florida Atlantic University

Theme: "Endless life cycles most beautiful: a celebration of the evolutionary and ecological diversity of parasite life histories".

3:00 **Sarah Goodnight**: Introduction

3:10 (48) **Scott Gardner**. PARASITES AND THE INTERCONNECTED BIOSPHERE.

3:35 (49) **F. Agustín Jiménez**. PARASITES WITH AGENCY: DIVERSITY AND DISTRIBUTION OF MONOXENOUS NEMATODES IN NEW WORLD.

4:00–5:00 pm Parasite Trivia! Salon II

6:00–7:00 pm	**Auction Preview**	**Rooftop Ballroom & Plaza Room**
7:00–9:00 pm	**32nd Annual ASP Student Auction**	**Rooftop Ballroom & Plaza Room**

Saturday, July 15

7:00–11:00 am	**Registration**	**Ballroom Foyer**
8:30–10:00 am	**Genomics & Molecular Biology I**	**Salon II**

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

Presiding: **D.C. Ghislaine Mayer,** Manhattan College

8:30 (50) † **Ryne W. Maness**, Charles D. Criscione. CHARACTERIZATION OF IMMUNE-RELATED GENES IN THE INVASIVE GEKKONID SPECIES, *HEMIDACTYLUS TURCICUS*.

8:45 (51) † **Chelsea S. Thorn**, Charles D. Criscione. DEVELOPMENT OF GENOMIC RESOURCES FOR THE GECKO TAPEWORM, *OOCHORISTICA JAVAENSIS.*

9:00 (52) † **Xuqing Chen**, Kimberly Reece, Jeffrey Shields. THE TRANSCRIPTOME PROFILES OF DIFFERENT LIFE HISTORY STAGES OF *HEMATODINIUM PEREZI* (DINOFLAGELLATA: SYNDINIALES) FROM THE BLUE CRAB (*CALLINECTES SAPIDUS*).

9:15 (53) † **Leah A. Owens**, Sagan Friant, Bruno Martorelli Di Genova, Laura J. Knoll, Monica Contreras, Oscar Noya, Maria G. Dominguez-Bello, Tony L. Goldberg. PARASITE ASSEMBLAGE CHARACTERIZATION WITH A NOVEL HOST SIGNAL REDUCTION METHOD AND METABARCODING PIPELINE.

9:30 (54) † **Mai Fahmy.** ASSESSING BIODIVERSITY IN MADAGASCAR WITH LEECH-DERIVED IDNA: METHODOLOGICAL ADVANCEMENTS AND ECOLOGICAL INSIGHTS.

9:45 (55) † **Mia M. Sanchez**, Rebecca Schwarz, Emerie Danson, Saraya AlSaffar, Charles Hauser, Daniel A. Gold. ISOLATING DISCRETE LIFE CYCLE STAGES OF A COCKROACH-INFECTING GREGARINE FOR GENE EXPRESSION ANALYSIS.

Saturday, July 15th

8:30–9:45 am Ecology & Evolution II Salon III

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

Presiding: **Anindo Choudhury,** St. Norbert College

8:30 (56) † **Corinne L. Conlon**, Krysten L. Schuler, Christopher M. Whipps. SURVEILLANCE FOR *ECHINOCOCCUS MULTILOCULARIS*, AN EMERGING ZOONOTIC PARASITE IN NEW YORK WILD CANID POPULATIONS.

8:45 (57) **Colin D. MacLeod**, Lien T. Luong. THE FRUIT FLY, *DROSOPHILA NIGROSPIRACULA*, EXHIBITS THREAT-SPECIFIC NON-CONSUMPTIVE EFFECTS IN RESPONSE TO PARASITE AND PREDATION RISK.

9:00 (58) † **Jenna M. Hulke**, Charles D. Criscione. DOWNSTREAM EFFECTS OF CHANGING LIFE CYCLE COMPLEXITY: TESTING THE MATING SYSTEM AND INBREEDING DEPRESSION AMONG NATURAL POPULATIONS OF THE TREMATODE *ALLOGLOSSIDIUM PROGENETICUM*.

9:15 (59) **Derek A. Zelmer.** PATTERNS OF ASYNCHRONY IN HELMINTH PARASITES OF CENTRARCHIDS IN J. STROM THURMOND LAKE, SOUTH CAROLINA.

9:30 (60) † **Gyrhaiss K. Kasembele**, Auguste C. Manda, Michiel W. P. Jorissen, Wilmien J. Luus-Powell, Willem J. Smit, Maarten P. M. Vanhove. ASSESSMENT OF ANTHROPOGENIC IMPACTS ON THE AQUATIC ENVIRONMENT AND BIODIVERSITY IN THE KATANGESE COPPERBELT AREA (DR CONGO): A PARASITOLOGICAL APPROACH.

9:45 (46) † **Caroline Liang**, Lien T. Luong. HISTORY OF PARASITE INFECTION SHAPES THE EXPRESSION AND MAGNITUDE OF NON-CONSUMPTIVE EFFECTS IN CACTOPHILIC FLIES.

10:00–10:30 am Coffee Break Ballroom Foyer

10:30 am–Noon Taxonomy, Systematics, & Phylogeny III Salon II

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

Presiding: **Veronica Mantovani Bueno,** University of Connecticut

10:30 (61) † **Matthew A. Walker**, Kurt Neubig, F. Agustín Jiménez. ILLUMINA SEQUENCE DATA AND MORPHOLOGICAL VARIABILITY LEND SUPPORT TO FURTHER DIVERSITY AMONG NORTH AMERICAN *GYRINICOLA* (NEMATODA: OXYUROIDEA).

10:45 (62) † **Haley R. Dutton**, Stephen A. Bullard, Anita M. Kelly. SUPPLEMENTAL DESCRIPTION AND PHYLOGENETICS OF *DRACOVERMIS OCCIDENTALIS* (DIGENEA: LIOLOPIDAE) INFECTING THE INTESTINE OF AN AMERICAN ALLIGATOR, *ALLIGATOR MISSISSIPPIENSIS* FROM THE MOBILE–TENSAW RIVER DELTA, ALABAMA, USA.

11:00 (63) † **Allison Bryant**, Matthew Bolek, Gabriel Langford. HIDING IN PLAIN SIGHT: WHAT CAN ENDOGENOUS DEVELOPMENT AND GENETIC SEQUENCING TELL US ABOUT COCCIDIA DIVERSITY IN LIZARDS?

11:15 (64) † **Melody Sly**, Debra T. Clopton, Richard E. Clopton. A NEW GREGARINE INFECTING THE DUNE BEETLE, *EDROTES VENTRICOSUS*, (COLEOPTERA: TENEBRIONIDAE) IN THE MOJAVE DESERT OF CALIFORNIA.

11:30 (65) † **Ali Z. Lira-Olguin,** Nina Heunis, Sonja Matthee, Conrad A. Matthee, Lance A. Durden, Jessica Light. MOLECULAR PHYLOGENETICS OF THE ECTOPARASITIC SUCKING LOUSE GENERA *HOPLOPLEURA* AND *POLYPLAX* (PHTHIRAPTERA: ANOPLURA, HOPLOPLEURIDAE, AND POLYPLACIDAE).

11:45 (66) **Nehemiah M. Rindoria**, George N. Morara, Willem J. Smit, Coret van Wyk, Nico J. Smit, and Wilmien J. Luus-Powell. *LAMPROGLENA CLEOPATRA* (COPEPODA: LERNAEIDAE) FROM *LABEO VICTORIANUS* (CYPRINIFORMES: CYPRINIDAE) WITH FIRST RIBOSOMAL AND MITOCHONDRIAL GENETIC DATA.

10:30 am–Noon Host-Parasite Interactions II Salon III

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

Presiding: **Maria G. Castillo,** New Mexico State University

10:30 (67) † **Jasmine A. Groves**, Dana Calhoun, Pieter Johnson, Jeffery Wilcox, John Michael Kinsella. DIVERSITY OF HELMINTHS IN CALIFORNIA BIRDS IN RELATION TO HOST TRAITS

10:45 (68) **Marin Milotic**, Alan J. Lymbery, Andrew R. C. Thompson, Leighton J. Thomas, Stephanie S. Godfrey. PARASITE COMMUNITIES OF CRITICALLY ENDANGERED HOSTS ARE INFLUENCED BY DIFFERENT SITES OF TRANSLOCATION AS REVEALED BY METABARCODING.

11:00 (69) † **Adriana M. Perrucci**, Kristin K. Herrmann, Charles R. Randklev. PARASITES OF FRESHWATER MUSSELS OF TEXAS AND THE EFFECT OF TREMATODE PARTHENITAE ON HOST REPRODUCTION AND MORTALITY.

11:15 (70) **Mariel L. Campbell**, Teresa J. Mayfield-Meyer, Dusty L. McDonald, Sara V. Brant, Jonathan L. Dunnum, Joseph A. Cook. PARASITES, PATHOGENS, AND THE EXTENDED SPECIMEN.

11:30 (71) † **Jackson R. Snyder**, John F. Shea, Chantal Nguyen, Ben Hanelt. ALTERATION OF BEHAVIOR IN AQUATIC SNAILS DUE TO LARVAL INFECTIONS OF *PARAGORDIUS VARIUS.*

11:45 (72) † **Cameron Hodinka**, Jillian Detwiler. BEHAVIOURAL CHANGES IN POTENTIAL HOST SNAILS (*STAGNICOLA ELODES*) DEPENDS ON ABIOTIC AND BIOTIC FACTORS.

Saturday, July 15th

Noon–1:00 pm Editorial Luncheon Pavilion II

1:15–2:30 pm Genomics, Molecular Biology II & Immunology Salon II

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

Presiding: **Judith Humphries,** Lawrence University

1:15 (73) **Kaylee S. Herzog**, Joseph R. Fauver. BENCHMARKING Q20+ ONT MINION LONG-READ SEQUENCE DATA FOR GENERATING REFERENCE-QUALITY GENOMES FOR PARASITIC nematodes.

1:30 (74) **Elisha Enabulele**, Roy N. Platt, Oluwaremilekun G. Ajakaye, Victor S. Njom, Marian Onwude-Agbugui, Christopher R. Pam, Tim Anderson. AN ARRAY OF PCR-RFLP MARKERS FOR DIFFERENTIATING BETWEEN SCHISTOSOMA HAEMATOBIUM AND S. BOVIS IN FIELD SETTINGS.

1:45 (75) Lijing Bu, Lijun Lu, Martina Laidemitt, Si-Ming Zhang, Martin Mutuku, Gerald Mkoji, Michelle Steinauer, and **Eric S. Loker**. A GENOME SEQUENCE FOR *BIOMPHALARIA PFEIFFERI,* THE MAJOR VECTOR SNAIL FOR THE HUMAN-INFECTING PARASITE *SCHISTOSOMA MANSONI.*

2:00 (76) † **Kayhan Ilbeigi**, Dorien Mabille, Mathieu Claes, Carl De Trez, Benoît Stijlemans, David Pérez-Morga, Guy Caljon. THE JOURNEY OF TSETSE TRANSMITTED TRYPANOSOMES: FROM THE SKIN INVASION TO A SYSTEMIC INFECTION.

2:15 (77) † **Deblina Misra**, Taylor Tuono, Harley Bendzus Mendoza, Matthew Pinch, Immo Hansen, Maria G. Castillo. AN ANALYSIS OF PHOSPHOPROTEOME NETWORKS IN RESISTANT (BS90) AND SUSCEPTIBLE (BB02) SNAILS OF *BIOMPHALARIA GLABRATA* SHOWS IMMUNOREGULATORY PATHWAYS.

1:15–2:30 pm Ecology & Evolution III Salon III

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

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

1:15 (78) † **Jennifer A. Talbert**, F. Agustín Jiménez, Andy W. Jones, Autumn J. Smith-Herron, Kristin K. Herrmann. HELMINTH COMMUNITIES IN MIGRATORY BIRD HOSTS OF THE FAMILY TURDIDAE.

1:30 (79) **Jeffrey A. Bell**, Laura E. Bell, Tyler J. Achatz, Vasyl V. Tkach. FEEDING GUILD DETERMINES HAEMOSPORIDIAN INFECTION RISK FOR WATERFOWL IN THE UPPER MIDWEST.

1:45 (80) Gina Magro, **Sarah A. Orlofske**, Jeffrey A. Bell, Vasyl V. Tkach, Bruce Urben, Robert C. Jadin. HAEMOSPORIDIAN DIVERSITY OF WATERFOWL IN GREEN BAY, WI.

2:00 (81) **Florian B. Reyda**, Brian Mullin. PECULIAR PATTERNS OF PARASITISM IN THE LAKE ONTARIO BASIN.

2:15 (82) **Sara V. Brant**, Eric S. Loker, Caitlin R. Babbitt, Martina R. Laidemitt, Erika T. Ebbs. PARASITES ARE EVERYWHERE, BUT ARE THEY IN MUSEUMS? MOVE FORWARD VOUCHERING PARASITE SPECIMENS IN THE 21ST CENTURY.

2:30–3:00 pm Coffee Break Ballroom Foyer

3:30–4:30 pm Student Business Meeting Salon III

Presiding: **Sarah Goodnight**, Florida Atlantic University

6:00–8:00 pm Poster Session Rooftop Ballroom & Plaza Room

† denotes student presentation in the Best Student Presentation Competition

ECOLOGY & EVOLUTION

83 **Janelle Laura J. Gacad**, Shoko Tanabe-Hosoi, Natalia I. Yurlova, Misako Urabe. TREMATODE SPECIES DETECTION AND QUANTIFICATION BY ENVIRONMENTAL DNA-QPCR ASSAY IN LAKE CHANY, RUSSIA.

84 **Katerina Sawickij,** Florian Reyda. PARASITES OF THE NORTHERN EVERGLADES, FLORIDA FISHES: BANGHAM, 1940 REVISITED.

85 **Hannah Whitcomb**, Florian Reyda. A SEARCH FOR THE MISSING FISH PARASITES: A SURVEY OF THE TRIBUTARIES OF ONEIDA LAKE, NEW YORK.

86 **Nicholas FitzGerald**, Emily Klawiter, John Shea. LINKING HORSEHAIR WORM PRESENCE WITH INSECT DIVERSITY: IMPLICATIONS FOR ECOSYSTEM HEALTH.

87 **Gavril L. Ibaan**, John Shea. EXTRAPOLATING LARGE SCALE PARASITE DATA FROM PUBLIC REPOSITORIES.

88 **Morgan Fleming**, Emily Bulmer, Florian Reyda. PARASITES OF COASTAL FISH OF GALVESTON BAY, TEXAS: CHANDLER, 1935 REVISITED.

89 **Maggie Bagatta**, **Ireland Coughlin**, **Rachel Horejsi**, Megan Wettstein, Jennifer E. Smith, Nicolas J. Wheeler. INTESTINAL HELMINTHS OF THE CALIFORNIA GROUND SQUIRREL (*OTOSPERMOPHILUS BEECHEYI*).

GENOMICS & MOLECULAR BIOLOGY

90 † **Shabnam Thakur**, Abhishek Mewara, J. S. Thakur, Rakesh Sehgal. GENETIC CHARACTERIZATION OF *CRYPTOSPORIDIUM* TO STUDY TRANSMISSION DYNAMICS IN NORTH INDIA.

91 **Katy A. Martin**, Andrew P. Jackson, Krysta McMahan, Jeba R. J. Jesudoss Chelladurai, Matthew T. Brewer. *TRITRICHOMONAS FOETUS* TRANSCRIPTOME: COMPARISON OF *IN VIVO* AND *IN VITRO* PROFILES.

HOST-PARASITE INTERACTIONS

92 **Merhawi Alemu Birhanu**, Girmay Dessalegn Tarekegn, Gessessew Bugssa Hailu. PREVALENCE OF INTESTINAL PARASITES AND ASSOCIATED RISK FACTORS AMONG INFLAMMATORY BOWEL DISEASE SUSPECTED PATIENTS IN TIGRAY REGIONAL STATE, NORTHERN ETHIOPIA.

93 † **Ester Shimon**, **Jamie E. Hungerbuhler**, Whitney C. Preisser. WHAT'S IN A FISH? A SURVEY OF THE PARASITIC FAUNA OF FOUR FRESHWATER SCULPIN (*COTTUS* SPP.) IN GEORGIA.

94 † **Sarah M. Martin**, Tori Guarino, Christina Anaya. TICKS RECOVERED FROM THE NONNATIVE BURMESE PYTHON (*PYTHON BIVITTATUS*) IN SOUTHWEST FLORIDA.

95 **Katy A. Martin**, Krysta McMahan, Jeba R. J. Jesudoss Chelladurai, Doug E. Jones, Matthew T. Brewer. NOVEL METHODOLOGY FOR THE STUDY OF *TOXOCARA CANIS* SOMATIC LARVAE.

96 **Julie Funchess**, Christina Anaya. AGGRESSION IN MALE HOUSE CRICKETS (*ACHETA DOMESTICUS*) INFECTED WITH HAIRWORMS.

97 **D.C. Ghislaine Mayer**. MOLECULAR CHARACTERIZATION OF HUMAN INTESTINAL PARASITES IN BIVALVES.

98 **Victoria C. Farrington**, John Shea. Viability of snail-to-snail transfer of *Paragordius varius* cysts to crickets.

99 **Luke Niemann**, Christina Anaya. REPORT OF PARASITES FROM THE GASTROINTESTINAL TRACT OF CANE TOADS (*RHINELLA MARINA*) IN SOUTHWEST FLORIDA.

100 † **Ryan W. Koch**, Kyle D. Gustafson, Matthew G. Bolek. OBSERVATIONS ON TURTLE ACANTHOCEPHALAN ABNORMALITIES FROM OKLAHOMA, U.S.A.

101 † **Md Khalilur Rahman**, Md Hasanuzzaman Talukder, Babul Chandra Roy, Hironmoy Biswas. PREVALENCE AND DIVERSITY OF GASTROINTESTINAL PARASITES ASSOCIATED WITH DIARRHEA IN BUFFALO CALVES UNDER EXTENSIVE BUFFALO FARMING IN BANGLADESH.

102 **Thomas B. Hayssen**, Noah Takamiya, James Pfaff, John Shea. *CHORDODES MORGANI* EFFECT ON HOST GROWTH AND ITS DEVELOPMENT TIME IN DIFFERING DIET AND TEMPERATURE CONDITIONS.

103 **Peter Halvarsson**, Jessica Lernå, Ylva Hedberg Alm, Eva Tydén. RISK FACTORS FOR *STRONGYLUS VULGARIS* INFECTIONS IN GRAZING HORSES IN SWEDEN.

LIFE CYCLES & EPIDEMIOLOGY

104 **Taiye Shade Olusegun-Joseph**, Daniel Akinsanya Olusegun-Joseph, Muinah Adenike Fowora, Akinlabi Mohammed Rufai, Monsuru A. Adeleke. CASE REPORT: SEVERE INFESTATION WITH TUNGIASIS IN A COASTAL COMMUNITY IN BADAGRY LAGOS, NIGERIA.

105 † **Ethan Over**, John Shea. ALTERNATIVE DEFINITIVE HOSTS FOR THE HORSEHAIR WORM *CHORDODES MORGANI*.

106 **Roger Ramirez-Barrios**, Anne Zajac, Elise O'Connell, Marko Estrada, Andy Plum, Joerg Balzer, Erica Miller, Kim Royar, Alexandra Lombard, Megan Kirchgessner. *ECHINOCOCCUS MULTILOCULARIS*: IS THE EUROPEAN HAPLOTYPE ENDEMIC IN THE EASTERN US?

107 **Gabriel J. Langford**, Maria Coppola, Andrea Eways. BIRDS CAN SERVE AS A DISPERSAL AGENT FOR *GYRINICOLA BATRACHIENSIS* (OXYUROIDEA: PHAYNGODONIDAE) THICK-SHELLED EGGS.

CHEMOTHERAPY & DRUG RESISTANCE

108 † **Anandi Goel**, Geetika Narang, Jahnvi Jakhan, Vineeta Singh, Monika Matlani. ARTEMISININ BASED COMBINATION THERAPY FAILURE IN COMPLICATED *PLASMODIUM FALCIPARUM* MALARIA WITH CONCURRENT ENTERIC FEVER: A CASE REPORT FROM NORTH INDIA.

TAXONOMY, SYSTEMATICS, & PHYLOGENY

109 Ryan P. Shannon, **Matthew G. Bolek**. A METHOD FOR ISOLATING AND SEQUENCING TRYPANOSOME CELLS TO INVESTIGATE SPECIES ASSOCIATIONS IN MULTIPLE MORPHOTYPE INFECTIONS.

110 † **Zoe S. Von Holten**, Tyler J. Achatz, Lynda R. Lafond, Jakson R. Martens, Vasyl V. Tkach. IMPROVING DIAGNOSTICS OF *CRASSIPHIALA BULBOGLOSSA*, THE TYPE-SPECIES OF *CRASSIPHIALA* AND A COMMON CAUSATIVE AGENT FOR BLACK SPOT DISEASE IN FISHES.

111 **Sabrina C. Sibrava**, Mariel Campbell, Joseph A Cook. SYSTEMATICS AND PHYLOGEOGRAPHY OF FLEAS IN THE SUBFAMILY DORATOPSYLLINAE (SIPHONAPTERA: HYSTRICHOPSYLLIDAE).

112 George Ezeokeke, Johnson Awoyele, **Nicholas J. Negovetich**. IMAGING PARASITES USING VARIABLE PRESSURE SCANNING ELECTRON MICROSCOPY.

113 † **Vincent T. Pickens**, Christina Anaya. A COMPARISON OF PARASITES IN THE DIGESTIVE TRACTS OF THE INVASIVE BURMESE PYTHON (*PYTHON BIVITTATUS*) COMPARED TO NATIVE SNAKES IN SOUTHWEST FLORIDA.

114 † **Gavin Godley**, Kirsten Jensen. RHINEBOTHRIIDEANS OF TWO SPECIES IN THE STINGRAY GENUS *HYPANUS.*

115 **Kayla Grey**, Katelyn Amspacher, Clayton Nielsen, F. Agustín Jiménez. OF INTESTINAL HELMINTHS AND *MEPHITIS MEPHITIS*: THE IMPORTANCE OF DNA BARCODING.

116 **Anindo Choudhury**, Carson Torhorst. PARASITES OF FISHES IN STREAMS OF SOBERANIA NATIONAL PARK, CHAGRES RIVER DRAINAGE, PANAMA.

117 † **Kara M. Heilemann**, Janine N. Caira. NO LONGER CHAMBERS OF SECRETS: CHARACTERIZING THE ANATOMY AND MUCOSAL SURFACE OF THE SPIRAL INTESTINES OF BATOID HOSTS BELONGING TO “TETRAPHYLLIDEAN” CESTODE CLADE 4.

IMMUNOLOGY

118 Justine N. Farrell, **Melissa K. Stuart**. *TRICHOMONAS TENAX* STIMULATES HL-60 NEUTROPHIL CHEMOTAXIS *IN VITRO.*

Sunday, July 16

8:00–10:00 am Insect Physiology & Behavior Symposium Salon II

Time (Abstract No.)

Presiding: **Julián Hillyer,** Vanderbilt University
Christina Anaya, Florida Gulf Coast University

8:00 **Julián Hillyer**. Introduction

8:10 (119) Collin Horn, Sarah Robinson, **Lien T. Luong**. ECOLOGY OF FEAR: ECTOPARASITES EXERT NON-CONSUMPTIVE EFFECTS ON HOST LARVAE WITH CONSEQUENCES FOR ADULTS.

8:25 (120) **Upendra R. Bhattarai**, Eddy Dowle, Robert Poulin, Neil J. Gemmell. DECODING THE MOLECULAR MECHANISMS OF HOST MANIPULATION BY PARASITES.

8:40 (121) **Jean-François Doherty**, Bingzong Liu, Ben Hanelt, Eddy J. Dowle, Neil J. Gemmell, Robert Poulin, Leonard J. Foster, Benjamin J. Matthews. TO “DRIVE” A LANDLUBBING INSECT INTO DEADLY WATERS: A HAIRWORM STORY.

8:55 (122) **Christina Anaya**, David Benhaim, Matthew G. Bolek. INFECTED CRICKETS EAT LESS AND CONVERT FOOD TO ENERGY LESS EFFICIENTLY THAN UNINFECTED CRICKETS.

9:10 (123) **Julián F. Hillyer**, Jordyn S. Barr, Tania Y. Estévez-Lao. THE SIZE AND BODY COMPOSITION OF MOSQUITOES IS ALTERED BY ENVIRONMENTAL TEMPERATURE AND AGING.

9:25 (124) **Alex P. Arp**. EMBRYONIC FEMALE-LETHAL NEW WORLD SCREWWORM STRAINS, HIGH EXPRESSION, LOW PERFORMANCE.

9:40 (125) **John F. Shea**, Amy M. Worthington. TESTING BEHAVIOR ALTERATION IN CRICKETS EXPERIMENTALLY INFECTED WITH HORSEHAIR WORMS.

10:00–10:30 am Coffee Break Ballroom Foyer

10:30 am–Noon Taxonomy, Systematics, & Phylogeny IV Salon II

Time (Abstract No.)

Presiding: **Florian Reyda,** State University of New York at Oneonta

10:30 (126) **Veronica Mantovani Bueno**, Bruna Trevisan, Janine N. Caira. HOW SURREAL ARE THE ESCHERBOTHRIIDAE? A MOLECULAR PERSPECTIVE.

10:45 (127) **Michael Tessler**, Spencer C. Galen, Rob DeSalle, Bernd Schierwater. NAMING CRYPTIC PARASITE SPECIES WITH MOLECULAR MORPHOLOGY.

11:00 (128) **Janine N. Caira**, Kirsten Jensen. THE MORE YOU KNOW THE BETTER THE ESTIMATE.

11:15 (129) **Stephen A. Bullard**, Micah B. Warren, John H. Brule. A NEW GENUS AND SPECIES OF CATHARIOTREMATINAE BULLARD, 2021 (MONOGENOIDEA: MONOCOTYLIDAE) INFECTING THE OLFACTORY LAMELLAE OF DEEPWATER GULPER SHARKS, *CENTROPHORUS GRANULOSUS* (SQUALIFORMES: CENTROPHORIDAE) IN THE GULF OF MEXICO.

11:30 (130) **Kenzie Grover**, Voitto Haukisalmi, Kurt Galbreath. THE MONGOLIAN REMODELING AND THE STRUCTURE OF HOST-PARASITE DIVERSITY ACROSS CENTRAL ASIA.

11:45 (131) **James P. Bernot**, Geoff A. Boxshall. TAXONOMY, SYSTEMATICS, AND NEW KEYS FOR CALIGUS (COPEPODA: CALIGIDAE) ILLUMINATED BY CONFOCAL LASER SCANNING MICROSCOPY

10:30 am–Noon Host-Parasite Interactions III Salon III

Time (Abstract No.)

Presiding: **Deblina Misra,** New Mexico State University

10:30 (132) **Matt Brewer**, Katy Martin, Krysta McMahan, Doug Jones. IMPLANT DEVICE THAT ENABLES IN VIVO STUDY OF HELMINTHS AND MIMICS PARASITE ANTIGEN LOAD RESULTING IN A NOVEL VACCINE STRATEGY.

10:45 (133) **Adebayo J. Molehin**, Brooke Hall, Christine Lee, Sean A. Gray, Darrick Carter. CHARACTERIZATION AND PROCESS DEVELOPMENT OF SERINE PROTEASE INHIBITOR (SJ-B6): A NEXT GENERATION TRANSMISSION-BLOCKING VETERINARY MRNA VACCINE FOR ASIATIC SCHISTOSOMIASIS.

11:00 (134) **Gina M. Gallego Lopez**, William J. Olson, Andres M. Tibabuzo Perdomo, David Stevenson, Daniel Amador-Noguez, Grant Lawrence, James W. Leahy, Laura Knoll. KISS AND SPIT METABOLOMICS HIGHLIGHTS THE ROLE OF HOST PURINE METABOLISM DURING PATHOGEN INFECTION.

11:15 (135) **Kevin S. Lievano**, Scott L. Gardner. DETECTION OF TRYPANOSOMATID PARASITES IN BEE ASSASSIN BUGS (*APIOMERUS* SPP.) IN KEITH COUNTY, NEBRASKA.

11:30 (136) Roger R. Barrios, Anne C. Jones, Mason C. Martin, Tappey H. Jones, **Ashleigh B. Smythe**. VOLATILES IN FECES OF RATS INFECTED WITH THE TAPEWORM *HYMENOLEPIS DIMINUTA.*

11:45 (137) **Matthew Bolek**, Maria Reid Bollinger, Taylor Michelle Fiedor, Kyle Douglas Gustafson. A REVIEW OF *PFEIFFERINELLA* (APICOMPLEXA: PFEIFFERINELLIDAE) INFECTIONS IN NORTH AMERICAN FRESHWATER GASTROPODS.

Sunday, July 16th

1:00–2:00 pm H.B. Ward Medal Lecture Salon II & III

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

1:00 pm **Jillian Detwiler**. Introduction of the 2023 H. B. Ward Medal Recipient.

1:10 pm **Charles Criscione**. "PEDIGREE OF A PARASITOLOGIST PROGENY."

Dr. Charles Criscione. Recipient of the 2023 H. B. Ward Medal.

2:15–4:30 pm ASP Awards & Business Meeting Salons II & III

ASP Awards: Distinguished Service Award

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

Dr. John Hawdon. Recipient of the Distinguished Service Award.

Sunday, July 16th

ASP Awards: Ashton Cuckler New Investigator Award

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

Dr. Jimmy Bernot. Recipient of the Ashton Cuckler New Investigator Award.

Best Student presentations and Marc Dresden Travel Grant Awards

Presiding: **F. Agustín Jiménez**, Southern Illinois University

ASP Business Meeting

Presiding: **Tamara J. Cook,** Sam Houston State University

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

See you all next year July 11–14 in Denver, CO!

Abstracts

1

Microfilaremia in sheltered canines infected with *Dirofilaria immitis* in the Cumberland Gap region of Tennessee, Kentucky, and Virginia.

Charles T. Faulkner, Crystal LeRoy
Lincoln Memorial University, Harrogate, TN, USA

Cases of canine heartworm (CHW) disease continue to increase in incidence annually despite efforts to increase awareness of its adverse impact and availability of effective and affordable prophylactic medications. Research in the Cumberland Gap region (CGR) has focused on the occurrence of CHW in the local pet dog and mosquito vector populations. Wild Canidae, primarily coyotes (*Canis latrans*), have also been surveyed opportunistically to assess their role as a source of microfilariae for transmission to suitable mosquito vectors. This study was undertaken to better estimate the contribution of stray and surrendered dogs resident in CGR shelters to CHW transmission in the regional vector population. Local shelters are visited weekly and whole blood samples (n=147) collected and tested for CHW antigen without immune complex disassociation by heat treatment. Additionally, canines obtained extra-locally (n=200) from regional shelters for use in the LMU-CVM surgery program were tested for CHW antigen and screened for microfilaremia. To date, a total of 347 dogs from all sources have been tested for CHW antigen with an estimated local prevalence of 7.5% and extra-local prevalence of 3.5%. Compared prevalence of CHW positivity was not significantly different by location (p = 0.140). Blood from CHW antigen positive dogs was examined microscopically for microfilariae. Circulating microfilaria (>30 um) of *Dirofilaria immitis* were observed in 8 (72.7%) dogs locally, and 5 (71.4%) extra-local dogs supporting their role as transmission sources for infection of the regional mosquito vector population.

2

Investigating digenean diversity in an estuarine pond adjacent to the Gulf of Mexico on the Fort Morgan Peninsula, Alabama.

Stephen S. Curran, John H. Brule, Steven P. Ksepka, Triet N. Truong, Micah B. Warren, Stephen A. Bullard
Auburn University, Auburn, AL, USA

We investigated the digenean community from a~ 5 acre estuarine pond adjacent to the southern portion of Mobile Bay and the northern Gulf of Mexico in Alabama (30°14′ 04.54″ N, 87°58′ 09.84″ W). Opportunistic collections of invertebrates and estuarine fishes were conducted on several occasions during the Spring of 2023. Digeneans were collected and preserved for morphological observation and DNA extraction. The internal transcribed spacer 2 region and a portion of the 28S ribosomal gene were sequenced from individual specimens. Preliminary identifications from collections revealed 7 adult digenean species (1 Haplosplanchnidae and 6 Haploporidae) infecting 5 species of fishes. Two metacercariae (*Rhipidocotyle lepisostei* and *Ascocotyle* sp.) infected *Menidia beryllina*, and the metacercaria of *Microphallus* cf. *turgidus* infected the grass shrimp *Palaemon pugio*. Cercariae representing at least 5 species (3 Haploporidae spp., 1 Heterophyidae sp., and 1 Microphalloidea sp.) developed in rediae infecting 4 hydrobiid snail species: *Littoridinops monroensis*, *Littoridinops palustris*, *Heleobops* sp., and *Pyrgophorus* cf. *platyrachis*. Digenean identifications will be reported to lowest possible taxonomic level. The most significant result thus far is the recovery of an undescribed species of *Saccocoelioides* (Haploporidae) having a cercaria that develops in *L. monroensis* and matures in *M. beryllina*. This species is identical to specimens originally discovered and identified as undescribed by

longtime ASP member William F. Font. Bill collected the worm from *M. beryllina* in tidal creeks emptying into Lake Pontchartrain in Louisiana in 2011.

3

***Sarcocystis* in wildlife from the Midwestern US.**

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Sarcocystis cause a range of diseases in domestic animals and wildlife. The most well-known species, *S. neurona*, causes neurological disease in horses. Numerous species of domestic animals, wildlife, and humans can act as definitive hosts or intermediate hosts and sometimes both. The geographic distribution of *Sarcocystis* is cosmopolitan. Surveys of wildlife for the presence of *Sarcocystis* are limited in the Midwestern US. In this study we examined three abundant wildlife species, raccoons (*Procyon lotor*), coyotes (*Canis latrans*), and Virginia opossums (*Didelphis virginiana*) for the presence of *Sarcocystis* in skeletal muscle tissue. We used histological techniques to morphologically identify species and molecular techniques to confirm identifications. Microscopically, we found *Sarcocystis* in each of the wildlife species sampled at similar prevalences of 37.5% in raccoons (6/16), 27.3% in coyotes (3/11), and 19% in opossums (6/16). Molecular screening and identification are currently being conducted. The morphology suggests that opossums are infected with *S. neurona*, however, the species we found in the raccoons and coyotes were not distinctive enough for identification. Our study shows that several wildlife species are infected with *Sarcocystis* in Illinois, and the Virginia opossum may be acting as both intermediate and definitive hosts for *S. neurona*.

4

Prevalence of malaria and its associated risk factors in selected malarious kebeles of Gindeberet District, Western Ethiopia: Community based cross-sectional study.

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Malaria is one of the major fatal diseases caused by *Plasmodium* parasites and spread by infected female *Anopheles* mosquitoes. It can be either asymptomatic or symptomatic. Based on the World Health Organization report of 2021 it is endemic in eighty-five countries and territories including sub-Saharan African countries. Data are scarce on the prevalence of malaria and its associated risk factors in the Gindeberet district. This study aimed to assess the prevalence of malaria and its associated risk factors in selected malarious kebeles of Gindeberet district communities, in western Ethiopia. The study comprised 845 study subjects residing in 211 households conducted from September to December 2021 in selected kebeles of the district. A multi-stage sampling technique was used to determine the kebeles, and a systematic random sampling technique was used to select households by using the lottery method from the malarious kebeles of the district. Blood film microscopy and rapid diagnostic test (RDT) were used to determine malaria infection. Data were analyzed using SPSS version 23 statistical software. Results: A total of 845 study subjects were included in the study, of which 803 study subjects were afebrile (<37.5co) and 42 were febrile (≥37.5co). From afebrile study subjects, 25 (3.1%) and 24 (2.98%) were positive for malaria by RDT and microscopy, respectively. Moreover, the prevalence of malaria was 11.9% (5/42) among individuals with fever in both methods of microscopy and RDT. The overall malaria prevalence was 30 (3.55%) by both methods. The majority of malaria cases (76.7%) were caused by *P. falciparum* and the remaining were due to *P. vivax*. In this study, no mixed infection was identified.

Malaria prevalence was significantly higher among males AOR=4.23 (1.73,10.20), those that stay outdoors during the night time AOR=2.90 (1.04,8.01), and axillary body temperature AOR=4.10 (1.33,12.54), however, those >grade 12 AOR=4.73 (1.80,9.72), those that used Insecticide-treated net (ITN) in the preceding night AOR= 4.01 (2.21,11.14), and education message about malaria from any source AOR= 2.94 (1.13,7.70) were negatively associated with malaria. Malaria is still a public health problem in district. Males, those with fever, nighttime outdoor activities were at higher risk of malaria; however, malaria prevalence was significantly lower in those that used ITN the preceding night and those with education messages about malaria from any source. The burden of malaria in the community needs to be further studied using more sensitive diagnostic tools.

5

Prevalence of *Neospora caninum* antibodies in Pakistani Camel breeds (*Camelus dromedarius*) and its effect on the abortive history of animals.

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Neospora caninum an obligatory coccidian parasite is considered as one of the major abortive pathogens in a wide variety of domestic as well as wild animals. The Camelidae family may also serve as intermediate host for *Neospora caninum*. The present research was conducted to evaluate the seroprevalence of *Neospora caninum* antibodies in the sera of various breeds of camel from Dera Ghazi Khan district, Pakistan. Serum samples from one hundred and eighty-four clinically healthy camel individuals (male=31, female=153) were collected and processed for the detection of antibodies to *N. caninum* by using a commercial Enzyme immunoassay (cELISA) diagnostic kit (ID Screen® *Neospora caninum* Competition, Garbels, France). Twenty-seven (14.67%) of 184 serum samples were detected as positive against anti-*Neospora caninum* antibodies. A significant ($P < 0.05$) difference in seropositivity was recorded between male (19.35%) and female (13.73%) individuals. We found the highest rate of prevalence to *N. caninum* in Gaddi breed 28.57% (6/21) followed by Brela 20.59% (7/34), Maracha 11.84% (9/76) and Gulmani 9.43% (5/53). However the prevalence rate was statistically non-significant ($P > 0.05$) among various breeds of camel. An increasing pattern of positivity against *N. caninum* was noted with the age of camel demonstrating the horizontal transmission of infection. The seroprevalence to *N. caninum* based on feeding management of camels (natural grazing, stall feeding, and mixed type including both grazing and stall feeding) was examined as a significant ($P < 0.05$) risk factor. The prevalence rate was significantly ($P < 0.05$) higher in female individuals with abortion history 18.05% (13/72) that animals have had no such history 9.8% (8/81). The occurrence of antibodies against *Neospora caninum* in Pakistani camel suggests the further studies to investigate its epidemiological role and economic impact on the camel industry.

6

ELISA based detection and prevalence of IgG antibodies to *Toxoplasma gondii* infection in clinically healthy pet dogs from South region of Punjab, Pakistan.

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Toxoplasma gondii can parasitize all the warm-blooded animals, including humans and dogs. It may cause severe adverse effects in immunocompromised individuals and pregnant female. Pet dogs act as

companion animals to human due to their friendly behavior, also act as important intermediate host for *Toxoplasma gondii*. A cross sectional study was designed to evaluate the prevalence of *Toxoplasma gondii* antibodies and epidemiological risk factors in 184 serum samples of pet dogs from south region of Punjab, Pakistan. Serum samples were assayed to detect *T. gondii* IgG antibodies by using commercially available indirect ELISA Kit (ID Screen Toxoplasmosis Indirect® ID-VET Company, France). The overall prevalence rate of *Toxoplasma gondii* in pet dogs was 16.84% (31/184). Female dogs were more infected (18.55%) that of male dogs (14.94%) with a significant difference ($P < 0.05$) of prevalence between two categories. Samples obtained from older dogs (≥ 2 years) were more likely to be seropositive than the samples from adult dogs (1 year – 2 years) and young ones (1 month -1 year). The bull dogs had the highest prevalence rate 23.07% (6/26) compared to Katha breed 20% (1/5), Labrador 17.85% (5/28), Gultair 15.78% (3/19), German shepherd 15.38% (12/78), Bullterrier 12.5% (1/8) and Rottweiler 11.11% (1/9) while none of the beauty dogs were found positive to *T. gondii* infection. The seropositivity based on breeds of dog has no significant ($P > 0.05$) difference among various dog breeds. However the vaccination status, feed type, housing, sex of pet owner and exposure to cats was found as significant ($P < 0.05$) risk factors in the occurrence of *Toxoplasma gondii* infection. The findings of the current study exhibited that dogs play a vital role in the epidemiology of *T. gondii* and maintenance infection within the environment.

7

Elucidating the influences of microhabitat on gastropods to predict hotspots of brainworm (*Parelaphostrongylus tenuis*) transmission.

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Brainworm (*Parelaphostrongylus tenuis*) is one of the parasites that may be contributing to moose (*Alces alces*) population decline across North America. Brainworm is transmitted to moose through the ingestion of gastropods infected with the L3 stage. Thus, gastropod infection, presence, and abundance within habitat types has been used to estimate the risk of brainworm transmission to moose. However, finer-scale aspects such as where and when moose encounter and consume infected gastropods within a habitat type remain unknown. We hypothesized that the presence and abundance of gastropod hosts would be affected by temporal variation in climate and soil chemistry. We predicted that soil chemistry would have the greatest influence, with gastropod presence and abundance increasing during late summer, in areas with neutral soil (pH = 7.0) that is high in calcium. From June-August 2022, gastropods were surveyed from three locations in Eastern Manitoba where moose have recently declined. Using transect sampling, we collected gastropods and measured aspects of their microhabitat including proportion of land cover type, humidity, temperature, soil moisture, pH, and calcium. In total, 110 gastropods were collected from seven species, four of which are known brainworm hosts. None of the collected gastropods were infected, which is consistent with results from similar surveys conducted in North American moose habitat. Preliminary statistical analysis revealed that gastropod presence varied spatially, with their occurrence being most likely in the easternmost collection site, which was predominantly composed of shrubland, exposed land, and coniferous forest, and as humidity increased from 65-100%. In addition, we will discuss how microhabitat influences gastropod abundance to further understand the temporal and spatial variation of gastropods. My results will indicate the microhabitat factors influencing when and where brainworm is transmitted to moose. This information can be used by conservation initiatives to prevent brainworm transmission from gastropods to moose by employing land management interventions to reduce the probability that the two hosts encounter each other.

8

The most extreme trematode soldier caste occurs in a human-infectious, invasive freshwater species.

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Some species of marine trematode form colonies with a division of labor in their first intermediate host mollusk, with a soldier caste serving to defend the colony from invasion by other trematodes. Here, we show that a human-infectious freshwater trematode, *Haplorchis pumilio*, has a soldier caste, that those soldiers exhibit the most extreme morphological specialization for defense yet observed for trematodes, and that those soldiers help explain the dominance of *H. pumilio* in trematode component communities. The pharynx of *H. pumilio* soldiers, used for attacking enemies, averages 5.0 times greater in absolute volume than the pharynx of reproductives. All previously reported soldiers have had a pharynx of equal or smaller absolute size than their reproductively mature clonemates. We also demonstrate that, unlike all previously documented trematodes with soldiers, *H. pumilio* soldiers seem incapable of transitioning to a reproductive state. Soldiers are clearly distinguishable from juvenile reproductives of the same size; in addition to larger pharynx, they have a longer gut, no brood chamber, and no germinal cells. From the above evidence, we posit that *H. pumilio* is the first trematode known to possess a sterile caste analogous to that of the social insects. *H. pumilio* soldiers aggressively attack other trematodes in its guild, while juveniles and reproductives do not attack. We show that, in part due to its effective defensive caste, *H. pumilio* appears to be a dominant competitor in the guild infecting the freshwater snail *Melanoides tuberculata* in its introduced range of the Americas. At the scale of individual lakes, we calculated *H. pumilio* is responsible for up to 100% of trematode deaths arising from competitive interactions between colonies. This species thus plays a major role in structuring parasite community composition. *H. pumilio* represents the first known freshwater trematode with a soldier caste, the first species with division of labor shown to dominate component community structure, and the first soldier caste that appears to be developmentally irreversible. As the adult stage of this trematode is recognized as pathogen of global importance, *H. pumilio* joins the ants, bees, wasps, and termites as a socially organized species that directly impacts humanity.

9

A widespread host-parasite association in the Strait of Georgia provides a unique opportunity for studying the effects of climate change on parasitism in the marine environment.

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Decades of research have clearly shown that the effects of climate change will substantially impact marine species through a variety of environmental factors, e.g. increase seawater temperature or decreased pH and salinity. In lab-based experiments, simulated climate change often elicits a species-specific response, highlighting the importance of research that investigates interspecific interactions, such as competition, predation, and parasitism. Parasitism is especially vulnerable to altered interspecific interactions, as the life cycle of many parasites cannot be completed without a particular combination of host species, i.e. obligate parasites. However, designing experiments that expose all species in a parasite's life cycle to multiple abiotic factors would be logistically challenging to say the least. One alternative to creating such nightmarishly complex experimental set-ups, is to use naturally

occurring systems that possess gradients in some or all of the abiotic parameters of interest, e.g. CO_2 vents that create pH gradients are used by ocean acidification researchers. This presentation describes my work on a trematode parasite found throughout the Strait of Georgia, British Columbia, and suggests that this body of water could be used as a natural laboratory to study host-parasite interactions in the context of climate change. *Maritrema gratiosum* (syn. *M. arenaria*) uses the acorn barnacle, *Balanus glandula*, as one of three host species to complete its life cycle. *Balanus glandula* is a ubiquitous component of intertidal ecosystems, and is abundant along naturally occurring gradients in pH, temperature, and salinity in the Strait of Georgia - the climate change hat-trick! Consequently, the distribution of *M. gratiosum* within barnacle host populations along these gradients may provide invaluable insight not only into how climate change will alter parasite dispersal, transmission, and development, but also how the combined effects of infection and climate change will impact host species from an individual to population level.

10

Parasite biodiversity in snakes from the U.S. and Canada.

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Snakes are widespread and diverse, but they are also difficult to survey, leaving many questions regarding their ecologies. In fact, snakes are one of the most understudied groups of vertebrate hosts. By consolidating snake–parasite studies, we hope to gain new insights into parasite biodiversity and snake ecology, including new inferences regarding difficult-to-observe trophic relationships. We conducted a systematic literature review of endoparasitic helminth and pentastome species that infect snakes in the U.S. and Canada. We used existing bibliographies and additional database searches to record information regarding snake hosts (e.g. species, size), parasites (e.g. species, prevalence), and habitats (e.g. region, aquatic vs terrestrial). We found that only 47% (83/175) of snake species have been examined for helminth and pentastome species. Among those, few have been surveyed extensively enough to capture at least 80% of their parasite communities. Most documented parasites were adults, and future morphological and genetic studies may elucidate the intermediate hosts for these parasites. Using sampling effort to estimate parasite species richness, we found that estimated parasite richness was higher in larger-bodied snake species and aquatic snake species. Aquatic snake species were among the best sampled taxa, and among four aquatic snake genera (*Agkistrodon*, *Farancia*, *Nerodia*, and *Regina*), most of the trematode species were unique to each snake genus (42% for *Agkistrodon*, 100% for *Farancia*, 51% for *Nerodia*, and 83% for *Regina*). Furthermore, most (9/13) of the aquatic snake species had at least one specialist trematode species that is not known to infect any other snake species in the U.S. or Canada. These results suggest that several described trematode species may be worth pursuing in the future as potential bioindicators of snake presence in aquatic ecosystems. If the life cycles of these trematodes can be elucidated, their presence in easily sampled invertebrate hosts may be useful indicators of more difficult to survey snake species. Based on the patterns observed here, we will recommend several future avenues for research where more extensive parasitological surveys of snakes and continued life cycle investigations could yield important ecological insights and benefit both snake and parasite conservation.

11

Worms got your tongue? Oral parasites alter male advertisement calls and disrupt expected relationships between male traits and female choice in green tree frogs.

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Frog tongueworms (*Halipegus occidualis*) are oral parasites that live in the mouths of many species of frog across North America; however, the impacts of infection on male advertisement calls are poorly understood. Single, calling male green tree frogs (*Dryophytes cinereus*) were recorded in the field, and visible parasites in the mouth were counted. Data on several call characteristics previously established as important for female choice were extracted using Audacity software and the R package warbleR. Infection intensity had significant impacts on dominant frequency (in kHz), call duration (in ms), and pulse structure of male calls. Specifically, heavily infected males had shorter calls with lower frequencies and lower pulse rates than mildly infected individuals. Pairwise call playback experiments were then conducted to determine if these observed differences in call characteristics affected female mate choice. Female frogs significantly selected the calls of uninfected males over heavily infected competitors, but surprisingly selected moderately infected male calls over uninfected male calls. Additionally, the presence of parasites in these pairwise mate choice decisions disrupted expected relationships between certain male traits (e.g., call frequency in kHz, body size) and female choice. Females selected higher frequency calls and smaller males more frequently when parasites were a factor in the decision, indicating that infection status may drive certain aspects of female choice in natural populations.

12

First taxonomically confirmed record of a gyrodactylid, *Gyrodactylus cichlidarum* Paperna, 1968 (Monogenoidea) infecting farmed Nile tilapia, *Oreochromis niloticus* (Linnaeus, 1758) (Cichliformes: Cichlidae) in the United States.

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The parasites of Nile tilapia, *Oreochromis niloticus* (Linnaeus, 1758) (Cichliformes: Cichlidae) are poorly documented in North America. These parasites concern fisheries managers and fish culturists in North America because they comprise potential introduced (exotic, non-native) pathogens that kill cultured tilapias and could theoretically harm sympatric wild fishes. As part of a survey of the parasites of Nile tilapia cultured at high density in flow-through raceways, we observed numerous monogenoid specimens attached to the skin, fins, and gill filaments that we identified as *Gyrodactylus cichlidarum* Paperna, 1968. This parasite was originally described from the skin and gill of mango tilapia, *Sarotherodon galilaeus* (Linnaeus, 1758) (Cichlidae) from pools and streams in the Accra plains, southwestern Ghana. Live specimens of *G. cichlidarum* intended for morphology were heat-killed, formalin-fixed, routinely stained, cleared, and whole-mounted on glass slides. Additional specimens were preserved in 95% ethanol for DNA extraction and sequencing of the internal transcribed spacers 1 and 2 (*ITS1*, I*TS2*) and *5.8S* ribosomal RNA gene. We identified our specimens as *G. cichlidarum* because they had a ventral bar with a distinct, dorsally-positioned postero-medial knob and marginal hooks with a wide aperture and flat sickle base. Our two identical sequences of the *ITS2-5.8S-ITS2* (783 bp and 723 bp) were nearly identical to (1 bp difference) one ascribed to *G. cichlidarum* from the United Kingdom.

The present study is the first taxonomically confirmed record of a parasite infecting Nile tilapia in the United States and comprises the first genetic information for *G. cichlidarum* in the United States.

13

Parasitological survey of two newly discovered populations of exotic invasive mystery snails (*Cipangopaludina*: Viviparidae: Bellamyinae) in Alabama and Arkansas.

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Mystery snails were introduced into North America in the early 20th century as food products from China and Japan and are common in the aquarium trade. Two populations of mystery snails (*Cipangopaludina* cf. *japonica*) were recently discovered in Arkansas (Lake Granada and Lake Estrella) and Alabama (Lewis Smith Lake), alarming fisheries managers concerned with potential snail-vectored pathogens and zoonotics. Because these recently discovered snail populations were un-surveyed for parasites, we collected and necropsied 464 and 213 snails from Arkansas and Alabama, respectively. Each snail was excised from its shell before each tissue was examined separately for parasitic infection using a stereo-dissecting microscope with bright and dark field stage illumination. Arkansas mystery snails were infected with encysted, unencapsulated metacercariae of *Alloglossidium* cf. *corti* (Digenea: Alloglossidiidae); which were removed from the epithelium of the visceral mass, excysted, observed with a compound microscope having differential interference contrast optics, heat-killed, preserved in formalin, stained in Van Cleave's and Ehrlich's hematoxylins, cleared in clove oil, and whole-mounted on glass slides. Additional specimens were preserved alive in 95% EtOH for DNA extraction and phylogenetics. These specimens were morphologically (having characteristic cercarial stylet) and genetically (nucleotide sequences for ITS2, 28S) similar to *Alloglossidium corti* (Lamont, 1921) Van Cleave and Mueller, 1934, which matures in the intestine of ictalurid catfishes. The snails from Arkansas were infected also by the 3rd or 4th stage larva (L3, L4) of *Spiroxys* sp. (Nematoda: Gnathostomatidae), which likely matures in the intestine of a snail-eating freshwater turtle. These specimens were 98–98.7% similar in the 18S to several *Spiroxys* spp. By those same methodologies, we found that Alabama mystery snails were infected by metacercariae of *Homalometron* sp. (Apocreadiidae) (confirmed with morphology and 28S nucleotide sequences) and another metacercaria having 28S and ITS2 sequences that were 100% and 99.8% identical to *Polylekithum ictaluri* (Allocreadiidae), respectively. We aim to surveil for additional infections in these and other mystery snail populations.

14

A new lineage of *Echinostoma* in the "*revolution* group" species complex transmitted by *Bulinus* snail in Nigeria.

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Echinostomes are a diverse and large group of trematodes (Echinostomatidae) whose definitive host includes fish, reptiles, birds, and mammals. The genus *Echinostoma* is the most important in public and veterinary health. The type species *E. revolutum* and several other species with 37 collar spines

constitute the "*revolutum* group" complex based on morphological or molecular data. In Africa, the diversity of echinostomes is understudied. While screening *Bulinus* snails for schistosome cercariae in the Wase Pandam wetland in central Nigeria, we recovered echinostomes from an individual snail. Partial fragments of the large subunit nuclear gene and mitochondrial Nad1 gene were sequenced for the echinostome isolates. Molecular phylogenetic analysis suggests that the novel data of the echinostome is a unique *Echinostoma* species within the "*revolutum* group." The echinostome isolate was within a clade comprising *Echinostoma sp*. IG, *E. revolutum* (*sensu stricto* and *sensu lato*), *E. pseudorobustum*, and *E. miyagawai*. We present the first molecular data of an echinostomatid from Nigeria and highlight the need for molecular prospecting of the trematode diversity in Africa. Future research will focus on elucidating the lifecycle of the echinostome and provide a morphological description of the adult for taxonomic classification.

15

Morphological description, life cycle, and phylogenetics of a new species of *Proterometra* (Digenea: Azygiidae) from the Cahaba River, Alabama, U.S.A.

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We herein describe a new species of *Proterometra* (Digenea: Azygiidae: Azygiinae) from the Cahaba River, Alabama, USA, and document that it asexually reproduces in the compact elimia, *Elimia showalteri* (Lea, 1860) (Cerithioidea: Pleuroceridae) and matures in the esophagus of the blackbanded darter, *Percina nigrofasciata* (Agassiz, 1854) (Perciformes: Percidae). Adults of the new species differ from congeners by having a small body and eggs having a wholly fimbriated surface that appears (colloquially) as a cilia-like brush border. Naturally-shed cercariae of the new species differ from those of its congeners by having a strongly claviform tail stem bearing aspinose mammillae, a single furca, excretory pores that open on the posterior margin of the single furca, and few eggs in the cercarial distome. The behavior of the cercaria further differentiates the new species. Live naturally-shed cercariae of the new species secrete a jelly-like adhesive that coats the surface of the furca and evidently facilitates attachment to the surface of glass, plastic, and snail shell. The attached cercariae vigorously wiggle and thrash about once attached, perhaps as if mimicking the larva of a stream insect so as to lure the blackbanded darter to eat it. Phylogenetic analyses recovered monophyletic Azygiidae, which comprised a monophyletic Leuceruthrinae Goldberger, 1911 and polyphyletic Azygiinae Lühe, 1909. The present study is the largest taxon sampling for Azygiidae and the first to include 28S sequences of a species of *Leuceruthrus*. Compact elimia and blackbanded darter are new host records for *Proterometra*. The new species is the 3rd congener reported from the Cahaba River, a region renowned for its fish and snail endemic biodiversity.

16

Nomasanguinicola dentata (Paperna, 1964) Warren and Bullard, 2023 (Digenea: Sanguinicolidae Poche, 1926) infecting African sharptooth catfish, Clarias gariepinus (Burchell, 1822) Teugles, 1982 (Siluriformes: Clariidae) from the Okavango River, Namibia: supplemental description and phylogenetic analysis.

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Fish blood flukes (Digenea: Chimaerohemecidae; Acipensericolidae; Sanguinicolidae; Elopicolidae; Aporocotylidae) comprise 175 species assigned to 46 genera, are occasional pathogens of cultured fishes, and are the ancestor to the schistosomes. Sanguinicolids infecting catfishes (Siluriformes) (10 species; *Nomasanguinicola* spp., *Plehniella* spp., monotypic *Cladocaecum*, monotypic *Kritsky*; *Sanguinicola chalmersi*) are morphologically and ecologically diverse. *Nomasanguinicola* spp. (*N*. *canthoensis* [type] infecting bighead catfish [Clariidae], *N*. *dentata*, and *N*. *clarias* infecting African sharptooth catfish) are unique by having denticles in two columns flanking the mouth. Like many freshwater fish blood fluke lineages, *N*. *dentata* and *N*. *clarias* have not been sequenced. Herein, we provide a supplemental description of *N*. *dentata* and reconstruct a 28S phylogeny. On 7 Dec 2021, we examined the heart of an African sharptooth catfish from the Okavango River (northeastern Namibia) that was infected with adults of *N*. *dentata*. Presently, no type-material exist for *N*. *dentata* (type locality Israel) nor *N*. *clarias* (Egypt). *Nomasanguinicola dentata* differs from *N*. *canthoensis* by having an intestine with 4 radial ceca and a testis with laterally directed lobes extending anterior to the intestine as well as lacking regularly spaced aggregates of tegumental papillae. We suspect that *N*. *dentata* and *N*. *clarias* are conspecific. The 28S sequence for *N*. *dentata* differed from *N*. *canthoensis* by 144 base pairs, and the phylogenetic analysis recovered the 2 species sister to one another. This is the first report of a fish blood fluke from sub-Saharan Africa, and the first report of a species of *Nomasanguinicola* in Africa in nearly 40 yrs.

17

Revealing diversity of the Diplostomidae (Digenea) parasitic in kingfishers.

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Kingfisher birds are definitive hosts to a diversity of digeneans that use fish as second intermediate hosts. Many of these digeneans belong to the family Diplostomidae. Their larvae are well-known causes of fish health problems throughout the world, including "black spot disease". Until recently, only a limited number of diplostomid species were known from kingfishers throughout the New World. In the present study, we studied the morphology and sequenced the large ribosomal subunit (28S) rDNA and cytochrome c oxidase 1 (cox1) mtDNA genes of 7 diplostomid taxa from kingfishers in Brazil, the USA, and the Philippines. Newly generated and previously published DNA sequences were used for phylogenetic analyses to explore the interrelationships of these digeneans and their close relatives. Two new species of the previously monotypic *Crassiphiala* were described. The expansion of the genus required amendment to the generic diagnosis. A new genus was erected for a former undescribed species-level lineage of *Crassiphiala*. Our study further demonstrated the hidden diversity of digeneans infecting kingfishers throughout the New World. This study was funded in part by the National Science

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A new species of *Ellipsomyxa* Køie, 2003 (Bivalvulida: Ceratomyxidae) infecting the gall bladder of *Pangasius macronema* Bleeker, 1850 (Siluriformes: Pangasiidae) from the Mekong Delta, Vietnam.

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During a survey of parasites infecting freshwater fishes in the Mekong River (Vietnam) during September 2018, disporic plasmodia containing myxospores consistent with the genus *Ellipsomyxa* Køie, 2003 (Bivalvulida: Ceratomyxidae) were observed in the gall bladder of 15 of 39 (38%) *Pangasius macronema* Bleeker, 1850 (Siluriformes: Pangasiidae). We used morphology and nucleotides (small subunit ribosomal DNA, SSU rDNA) to characterize the new species. We diagnose the new species as: myxospore ellipsoid, 12.0–15.0 (mean ± SD = 13.3 ± 0.9; N = 47) long, 8.0–9.0 (8.4 ± 0.5; 42) wide, 8.0–9.0 (8.5 ± 0.5; 11) thick, 2.0–5.0 (3.3 ± 0.8; 41) between polar capsules; polar capsules equal, subspherical, 3.0–4.0 (3.7 ± 0.5; 86) long, 3.0–4.0 (3.6 ± 0.5; 86) wide, with 5–7 polar tubule coils. The new species differs from all congeners by myxospore dimensions, polar tubule coil count, and SSU rDNA sequence data, and is morphologically most similar to *Ellipsomyxa adlardi* Whipps and Font, 2013, infecting the naked goby (*Gobiosoma bosc* [Lacepede, 1800] [Gobiiformes: Gobiidae]) in Lake Pontchartrain, Louisiana. The new species differs from *E. adlardi* by having a longer myxospore (12.0–15.0 [13.3] vs. 11.3–14.4 [12.4]) and having shorter polar capsules (3.0–4.0 [3.7] vs. 3.9–4.9 [4.3]). A phylogenetic analysis of the SSU rDNA including all published *Ellipsomyxa* sequences recovered the new species sister to *Ellipsomyxa ariusi* Chandran, Zacharia, Sathianandan, and Sanil, 2020 and *Ellipsomyxa* sp. (MK561979), both collected from threadfin sea catfish (*Arius arius* [Hamilton, 1822] [Siluriformes: Ariidae]) in India. Consistent with previous phylogenetic analyses of *Ellipsomyxa* spp., *Ellipsomyxa* was recovered as monophyletic. The new species is the first *Ellipsomyxa* reported from a freshwater fish in Asia and the first myxozoan reported from *P. macronema*.

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Morphological and molecular description of a *Crepidostomum* sp. (Trematoda: Allocreadiidae) and the diversity of *Crepidostomum* from the Pacific Northwest.

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The genus *Crepidostomum* (Trematoda: Allocreadiidae) has recently become a subject of taxonomic revisions and newly described taxa. Studies from Europe and Northeast Asia have described several new species (*C. brinkmani*, *C. pseudofarionis*, *C. oschmarini*, *C. achmerovi)*, suggesting expanded diversity within a species complex. These advances in charting the diversity of the genus have relied increasingly on molecular data for species description and delineation because the morphological differences are often small. In the Pacific Northwest two typical salmonid specialists, *Crepidostomum farionis* and *C. metoecus* (Caira, 1985) have been widely reported, both with an assumed holarctic distribution across several salmonid host species. A lack of molecular data from these two species in the Pacific Northwest prompted the use of an integrative taxonomic approach to understand the true diversity of *Crepidostomum* in this region. A survey of Mountain Whitefish (*Prosopium williamsoni*) from the Deschutes River Basin, Oregon, yielded gravid individuals of a *Crepidostomum* species from the gallbladder. The species from Mountain Whitefish has an identical site of infection as *C. wikgreni* from whitefish in Europe, but has a different egg size, which is a defining morphological characteristic for *C. wikgreni*. The species from Mountain Whitefish groups within the *C. farionis* clade for all three genes analyzed (28S, ITS, and CO1). It is distinct from *C. farionis* in having a more tubular body, larger, asymmetrical oral papillae, a differently shaped ventral sucker, and a different distribution of vitelline follicles. Accounting for the differences in host specificity, morphology, and genetics, we conclude that the Mountain Whitefish form is a new species of *Crepidostomum* and that the diversity of *Crepidostomum* in Salmonids is greater than originally described.

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In vitro **activity and mechanisms of actions of methanolic extract of *Trametes versicolor* (Turkey-Tail) mushroom against *Toxoplasma gondii.***

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Toxoplasma gondii (*T. gondii*) is an intracellular coccidian protozoan parasite that infects approximately 1/3rd of the global human population and results in severe disease in immunocompromised individuals, pregnant women, and fetuses. It also causes stillbirth, miscarriage, and birth defects in livestock animals leading to huge economic losses. Currently available anti-*T. gondii* drugs are either associated with high costs, undesirable side effects, and ineffectiveness to all groups of patients, as well as to all forms of parasites, or they are constrained by geographical barriers in terms of prescription and use. This study assessed the in vitro safety of the methanolic extract of Turkey-Tail (TT), higher fungi, on human foreskin fibroblast (HFF) cells and the inhibitory activity of TT extract against tachyzoites of *T. gondii* RH-RFP type I strain. As the natural products are usually anti-oxidant in nature, biochemical assays for the mitochondrial function of tachyzoites upon TT treatment were also performed. The TT extract was not that cytotoxic, with the 50% cytotoxic concentration (CC50) value to be >100 µg/mL and it inhibited the tachyzoites with a 50% minimum inhibitory concentration (IC50) of 5.98 ± 1.22 µg/mL, generating a selectivity index (SI) of >17.00. It was discovered that TT extract induced the production of both mitochondria superoxide and reactive oxygen species, and disrupted the mitochondria membrane potential (MMP) in *T. gondii* tachyzoites. Since TT extract was non-cytotoxic, even at higher concentrations, and inhibitory against *T. gondii* with a higher SI value, the extract could be a promising source of new compounds for the future development of anti-*T. gondii* drugs.

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The anthelmintic activity of praziquantel analogs correlates with different structure-activity relationships at TRPM$_{PZQ}$ orthologs.

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The anthelmintic drug praziquantel (PZQ), discovered in the 1970s, remains the mainstay for treatment of parasitic flatworm infections. After ~40 years of clinical usage, a putative target for PZQ was identified: a transient receptor potential ion channel in the melastatin subfamily (TRPM$_{PZQ}$). Intriguingly, it was previously reported that certain PZQ derivatives show differential activities between different species of parasites in vivo and ex vivo. For example, some PZQ analogs are more active against cestodes than against schistosomes. Here, we interrogated whether the different activities of PZQ analogs against these different parasites are also reflected in unique structure-activity relationships (SARs) at the $\mathrm{TRPM}_{\mathrm{PZQ}}$ orthologs found in these different organisms. To this end, multiple PZQ analogs were synthesized and functionally profiled against schistosome and cestode $\mathrm{TRPM}_{\mathrm{PZQ}}$. Functional profiling of these PZQ derivatives showed that 'the glove fits': SAR of molecules ex vivo on worms correlated well with the SAR of these molecules at their different $\mathrm{TRPM}_{\mathrm{PZQ}}$ orthologs. These data further support $\mathrm{TRPM}_{\mathrm{PZQ}}$ serving as the relevant therapeutic target of PZQ in parasitic flatworms.

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Natural variation in a parasitic flatworm ion channel underpins differential sensitivity of parasites to praziquantel.

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The drug praziquantel (PZQ) is the primary treatment for infections caused by parasitic flatworms. A target for PZQ was recently identified in schistosomes, a transient receptor potential ion channel in the melastatin subfamily ($\mathrm{TRPM}_{\mathrm{PZQ}}$); however, little is known about the properties of $\mathrm{TRPM}_{\mathrm{PZQ}}$ in other flatworms. $\mathrm{TRPM}_{\mathrm{PZQ}}$ orthologs were scrutinized in all currently available parasitic flatworm genomes and functionally profiled; $\mathrm{TRPM}_{\mathrm{PZQ}}$ is present in all parasitic flatworms, and the consensus PZQ binding site is well conserved. There were three loci of variation identified across the parasitic flatworm $\mathrm{TRPM}_{\mathrm{PZQ}}$ pocketome, including an acidic residue in the TRP domain. This residue acts as a gatekeeper, impacting PZQ residency within the $\mathrm{TRPM}_{\mathrm{PZQ}}$ ligand binding pocket. Functional profiling of trematode and cestode $\mathrm{TRPM}_{\mathrm{PZQ}}$ orthologs revealed differing sensitives to PZQ, matching the varied sensitivities documented clinically. In trematodes and cyclophyllidean cestodes, which display high sensitivity to PZQ, the gatekeeper TRP domain residue is an aspartic acid, allowing for nanomolar activation by PZQ. However, the presence of a glutamic acid residue, found in pseudophyllidean cestode $\mathrm{TRPM}_{\mathrm{PZQ}}$, was associated with lower PZQ potency. Functional profiling of a pseudophyllidean channel revealed micromolar potency of PZQ at *Spirometra erinaceieuropaei* $\mathrm{TRPM}_{\mathrm{PZQ}}$. The definition of these different binding pocket architectures explains why PZQ shows high therapeutic efficacy against specific fluke and tapeworm infections but is poorly effective against others. Effort to identify new therapeutics that tolerate this TRP domain residue would be immensely valuable if analogous variation were to ever occur in natural flatworm populations, and for the development of targeted therapies towards specific infections.

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Discovery of a novel therapeutic candidate against animal trypanosomiasis.

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Animal trypanosomiasis (AT) is a widespread disease caused by *Trypanosoma* spp. and has a devastating effect on animal husbandry all over the world due to the scarcity of efficient drugs and development of drug resistance, hence emphasizing the need for novel treatment options. Following previous identification of 3’-deoxytubercidin as a highly potent trypanocide with curative activity in mouse models of both stage-1 and stage-2 Human African Trypanosomiasis (HAT), we now present a comprehensive preclinical evaluation of new 6-amino substituted tubercidin analogues with promising activity against a broad range of AT species. Potent hits were identified *in vitro* across all important AT species, i.e. *Trypanosoma brucei brucei*, sensitive and isometamidium (ISM)-resistant *Trypanosoma congolense*, *Trypanosoma vivax*, *Trypanosoma evansi* (type A and B) and *Trypanosoma equiperdum*. Selected ‘hits’ were further tested for *in vitro* metabolic stability (using bovine, horse and piglet liver microsomes), *in vivo* mouse models for each AT species, genotoxicity assays and mode-of-action studies (i.e. genome-wide RNA interference library screening, metabolomics). Analogue 3 was highly active in *T. vivax*, *T. congolense*, *T. equiperdum*, *T. evansi* and *T. brucei* curative mouse models. Furthermore, there was no indication of *in vitro* genotoxicity as confirmed by Vitotox®, the micronucleus and the comet assays. Mode-of-action studies for 3 revealed that the P1 nucleoside transporter and adenosine kinase are involved in drug uptake and activation, respectively. Given the preferred target product profile for a broad-spectrum drug against AT, analogue 3 represents a promising ‘lead’ candidate for treatment of animal trypanosomiasis, regardless of the causative species.

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Altered activation response associated with multidrug resistance in the canine hookworm *Ancylostoma caninum*.

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Nearly 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 is used frequently due to high rates of reinfection. Although effective, this places significant selective pressure on parasitic nematodes to develop resistance. Resistance to benzimidazoles, macrocyclic lactones, and pyrantel in the canine hookworm *Ancylostoma caninum* has been previously documented. Here we report the characterization of a new, naturally occurring triple-resistant isolate of *A. caninum*. Using *in vitro* egg hatch, larval development, and larval feeding assays, we show that this isolate is resistant to the benzimidazole thiabendazole, the macrocyclic lactones ivermectin and moxidectin, and the nicotinic receptor agonist pyrantel. The isolate harbors a non-synonymous mutation in the codon encoding amino acid 167 of the ß-tubulin gene, which has been linked to benzimidazole resistance. Quantitative PCR indicated that the frequency of the resistant allele in the isolate was approximately 75%. Using an *in vitro* activation assay, we found that L3 of the isolate activate to lower levels than L3 from a susceptible strain and a strain resistant to thiabendazole and ivermectin in response to serum filtrate and S-methyl-glutathione. Larvae from the resistant isolate do not activate differently from other strains in the presence of cGMP, indicating the activation is inhibited upstream of the cGMP step in the activation pathway. Understanding the mechanisms of anthelmintic resistance is important for control strategies, efficient anthelmintic utilization, and monitoring emerging resistance.

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Caffeic acid potentiates pyrimethamine activity *in vitro.*

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Toxoplasma gondii is a zoonotic parasite that causes ocular and congenital defects in humans globally. Caffeic acid (CA) is one of the major polyphenolic acids often found in tea. CA has been reported to have both biological and pharmacological properties (e.g., antimalarial and antibacterial activities). However, little is known about its combination with pyrimethamine (PY) inhibitory activity against *Toxoplasma gondii* (*T. gondii*) *in vitro*. We reported for the first time the combination of CA and PY against T. gondii RH-RFP, a Type I strain tachyzoite at 72 hours of interaction. CA, PY, CA: PY (1:1), CA: PY (1:2), CA: PY (2:1) inhibited *T. gondii* tachyzoite growth with IC50s values of 1.01, 0.64, 0.16, 0.71, and 12.80 µM at 72 h, respectively. Noteworthy, in this study, we discovered that CA: PY (1:1) was synergistic, while CA: PY (1:2) was calculated to be additive, and CA: PY (2:1) was found to be antagonistic. The fractional inhibitory concentration index values were 0.48, 1.8, and 31.88 for CA: PY (1:1), CA: PY (1:2), and CA: PY (2:1) combinations, respectively. Interestingly, all these combinations were found not to be cytotoxic. CA, PY, and CA v PY combinations target the mitochondrion of *T. gondii* RH-RFP via elevated production of ROS, Superoxide, and disruption of mitochondrial membrane potential. Taking all these findings together, we believe that future experiments are needed *in vivo* to validate the efficacy and safety of the CA: PY (1:1) ratio as a future combination agent against toxoplasmosis.

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Persistence of antigens of Histidine-Rich Protein II and III after treatment with malaria drugs.

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Malaria is a life threatening disease mostly spread to people through bites of infected female *Anopheles* mosquitoes. A prompt and accurate parasitological confirmation by microscopy or alternatively by Rapid Diagnostic Tests (RDTs) is recommended by the WHO, prior to treatment. Antigen-detecting RDTs have been used for detection of malaria at the point of care, especially in malaria endemic countries. The antigens identified by RDTs have been reported to show varying persistence for weeks after clinical symptoms of malaria have disappeared and parasites apparently cleared from the host. This work is aimed at studying the duration of persistence of these antigens after malaria treatment with Sulfadoxine-Pyrimethamine and Artemisinin based combination in children and adult. Outpatient attending three different clinics were diagnosed for malaria using Histidine-Rich Protein II (HRPII) and Histidine-Rich Protein III (HRPIII). A total of 360 patients, that were positive for malaria were selected for the study. They were treated with Sulfadoxine-Pyrimethamine and Artemisinin-Combination Therapy (ACT) and were monitored for 30 days. The two RDTs kits were used to test for the presence of malaria parasites at interval of 5-9, 10 -14, 15-19 and 20 -24 post treatment. The antigens were persistent in the different age groups sampled after 1 to 9 days post treatment at varying degrees of 90 to 100% detection. The detection of the antigens started reducing after 10 - 14 and 15-19 days post infection, at an average of 47.7 to 32.7% and 3.5 to 1%, respectively. After 20 -24 days post treatment, the antigens were not detected. It was observed generally that the antigen of HRPIII cleared faster than HPRII after treatment. The antigens remained positive in varying amount of time in the patients after treatment. The positive results after antimalarial treatment based on the findings of this work, might not necessary be due to the presence of malaria parasite but could be as a result of the presence of RDTs antigens used. Caution should be taken when using RDTs diagnostic results for treatment of malaria, particularly after the patient has recently been treated.

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Parasitology teaching and research at small, undergraduate institutions: benefits and challenges.

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There are benefits and challenges involving parasitology teaching and research at primarily undergraduate institutions (PUIs). Teaching parasitology provides a unique experience for undergraduates and generates interest in students from interdisciplinary fields including medicine, pre-veterinary medicine, zoology, immunology, microbiology, and public health. A course in parasitology can provide an introduction into the complexities of our field and often provides the first exposure of students to the importance of parasitic organisms. The challenge seems to lie in finding a balance between medically relevant parasites and the study of parasites as a general discipline regardless of medical significance, as well as convincing colleagues of the virtues of a parasitology course. Research projects in PUIs can span a diverse set of possibilities as there are rarely limits on what can and cannot be investigated due to the absence of larger research grant obligations. Thus, our students can work on a variety of projects that can range from ones designed to fit with my own long-term research to ones tailored more to a student's specific interests. The most common approach that I use involves starting students working on parasite surveys and eventually specializing on a particular problem involving systematics, morphology, life cycles, or ecology of one or more parasites discovered in a survey. Projects have involved local wild vertebrates, invertebrates including insects, and specimens from older collections. I will describe some of the projects and will discuss the inherent challenges of undergraduate research including the short duration of a student's presence in a research setting, time limitations, funding constraints, and organizational hurdles. I will also describe how I try to overcome these challenges.

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Learning should be infectious: Uniting teaching and research in parasite ecology and evolution.

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Parasitology is a diverse field and the roles and responsibilities of parasitologists are as varied and complex as the organisms we study. These different career demands represent trade-offs in decisions of how to invest time and resources. These trade-offs become even more pronounced in environments with limited resources such as funding, equipment, and other institutional support, which is often the case at primarily undergraduate institutions. Using life history theory as an analogy for the academic career path and the variety of professional choices that shape investments in the key areas of teaching, research, and service, I will explore potential opportunities to optimize productivity, efficiency, and effectiveness in the environment of a primarily undergraduate institution. These trade-offs are not unique to parasitology, but parasitologists have some additional opportunities and constraints to consider. The trade-off between teaching and research can be addressed by integrating the two in several ways. First, converting your primary research into teaching activities helps you see your research in a new way and helps you recruit potential undergraduate researchers. Parasitology themed teaching activities are relatively rare so by publishing your teaching modules you improve teaching parasitology beyond your classroom. Second, Course-Based Undergraduate Research Experiences incorporate your research directly into your classroom. This method of teaching improves learning and helps student develop a scientific identity. Accumulating evidence demonstrates a positive impact of CUREs on faculty

research. Few students start college with the intention of becoming parasitologists so teaching materials about parasites can spark an interest in the field. Another trade-off exists between mentoring student research and protecting your time to complete your research. Meaningful contributions of undergraduates to research can be maximized through strategies for recruiting, training, and supervising. Even with the best strategies, turnover of undergraduates is frequent, often before projects are completed. Maintaining project continuity involves museum-level curation of samples, data, and records. These strategies can be applied to any stage of career or academic environment and can be improved over time. By documenting innovations and publishing those results in research or teaching journals we can make parasitology an attractive field to future researchers and teachers.

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Getting and keeping students hooked on fish parasite research.

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Survey work has been the foundation for the research program I developed at SUNY Oneonta, a primarily undergraduate institution (PUI). The location, central east New York, has great access to nature, with countless streams and lakes, including Otsego Lake, the location of our field station. The SUNY Oneonta Biological Field Station, which has bench space, boats and vans set the stage for the work. Funding was available in small portions from an internal grant program supported by a strong network of alumni. But the most important element for success was a large population of students who were hungry for hands-on experience. Students were lured to the lab with the promise of field work experience (collecting fish). Later, they grew to appreciate the bigger component of the work—the laboratory. Most of the 85+ students stayed in the lab for more than one semester. The central question of the survey was simple. What parasites are present in the fishes of Otsego Lake? A second question followed: What can we learn about the lake ecosystem from the presence of the parasites? Through research activities students developed a skill set that they could take with them for their career paths—field skills, observation, dissection, parasite preservation, preparation of stained whole mount slides, and ultimately identification via light microscopy. Everyone in the lab contributed to that survey through fish examination, specimen processing and databasing. Students who stayed longer took on independent projects to address more specific questions about particular species, focusing on morphology, including taxonomic problems, infection patterns, or pathology. As a result of the survey I was able to incorporate research into coursework, providing students with images and examples of locally-occurring parasites. These examples helped draw attention to the research and attract students, but the biggest recruitment tool was the other students themselves, and my colleagues in the Biology Department. While my example of research at a PUI here is specific, I suggest that a survey-centered research approach is one that can be done just about anywhere where there are ecosystems, students, and the motivation to build a long-term dataset.

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Research from day one: Engaging first-year students in research @ PUI's.

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Involving students in collaborative and original research has proven to be a powerful educational tool to attract, retain and engage students in STEM. Parasitology, by its very nature, is an exciting and integrative discipline that can fuel interest in STEM education, careers, and higher degrees. As inculcation of enthusiasm is one of the key elements in generating interest in STEM, greater attention should be given to fostering STEM interests at PUI's for freshmen, who are excited to begin their research journey from day one. Since 2005, the Marine Science program at Eckerd College developed and incorporated the Freshman Research Program (FRP) into its curriculum to offer opportunities for first year students to work closely with a faculty mentor on a research project during the academic year. Research topics vary across the fields of Marine Science, ranging from marine parasitology to ocean acidification to coastal geology. Since the program's inception, it has created a social infrastructure in which former FRP students become peer mentors and promoted an institutional research culture that has spread to other STEM and non-STEM majors. By engaging students in research from day one, FRP alumni have enhanced success in securing professional opportunities. Furthermore, students are better prepared to develop authentic independent research projects during their junior and senior years, and beyond. Indeed, students trained in parasitology research from day one have been successful in developing productive careers in STEM. I will address how to incorporate a FRP as part of teaching, mentoring and research responsibilities; the importance of involving different campus constituencies in establishing a FRP; and how the FRP experience can impact student success in STEM, particularly in Parasitology.

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Flaps, spikes, and tentacles: diversity of lecanicephalidean tapeworms parasitizing cowtail rays.

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Stingrays in the genus *Pastinachus* are host to a great diversity of tapeworms including members of the orders Diphyllidea, Lecanicephalidea, Onchoproteocephalidea, Rhinebothriidea, Trypanorhyncha, and "Tetraphyllidea." While as many as 33 lecanicephalidean species names have been proposed and reported to infect species of *Pastinachus*, only five species total in the genera *Anthemobothrium*, *Flapocephalus*, and *Tetragonocephalum* are currently considered valid. Our goal was to characterize the diversity of lecanicephalidean tapeworms parasitizing members of *Pastinachus* with a particular emphasis on new species and morphological novelty. A total of 35 stingray specimens representing three of the five recognized species of *Pastinachus* (*Pastinachus ater*, *Pastinachus gracilicaudus*, and *Pastinachus solocirostris*), and an additional undescribed species, collected from 10 localities throughout the Indo-Pacific region were examined for tapeworms. Lecanicephalideans encountered were prepared for light and scanning electron microscopy for morphological work and, when possible, molecular sequencing of the 28S rRNA gene. Based on morphology, representatives of the Lecanicephalidea found parasitizing *Pastinachus* consisted of multiple species in each of the genera *Anteropora* and *Flapocephalus*, as well as members of tentacle-bearing taxa, all in the family Polypocephalidae. Specifically, we identified 18 species: four species of *Anteropora*, as well as three species in a clade allied with *Anteropora* yet inconsistent with our current concept of the genus to varying degrees; four species of *Flapocephalus* (the two known and two new species); and seven species with apical organs in the form of tentacles. Fifteen of the 18 species appear to be new to science. Unique features or combinations of features necessitate emendation of the generic diagnoses of *Anteropora* and *Flapocephalus*. The species with tentacles are consistent with the generic diagnosis of either *Anthemobothrium* or *Polypocephalus*. However, generic assignment of these species with tentacles is complicated. Preliminary molecular data suggest the genus *Polypocephalus* to be non-monophyletic with respect to at least five other polypocephalid genera, and the taxa parasitizing *Pastinachus* to be composed of up to three independent lineages. Interestingly, specimens of *Tetragonocephalum* were not observed in the specimen of *Pastinachus* examined.

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Interesting results from the "Hotel California": On the species of *Caulobothrium* parasitizing stingrays in the eastern Pacific Ocean.

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The genus *Caulobothrium* remains one of the most poorly studied genera of cestodes parasitizing batoids. In recent years, the number of known species has expanded from 10 to ~30 species with the discovery of substantial undescribed novelty in previously unexamined groups of stingrays. An interesting trend has emerged within *Caulobothrium* in which multiple morphologically distinct species have been found parasitizing the same host species. In this study, we focused on the taxa found parasitizing two species of stingrays from the eastern Pacific Ocean—the bat eagle ray, *Myliobatis californica*, from the Gulf of California, Mexico and the Tumbes round stingray, *Urobatis tumbesensis*, from off coastal Ecuador. At present only two valid species of *Caulobothrium* are known from *M. californica* and none have been reported from *U. tumbesensis*. A subset of cestode specimens preserved in formalin from each host examined was prepared for and examined with light and in some cases also scanning electron microscopy. Sequence data were generated for a portion of the 28S rDNA gene for the species for which material preserved in ethanol was available. A Maximum Likelihood analysis was conducted on these data and data available for species of *Caulobothrium* collected from other localities throughout the world. *Myliobatis californica* was found to host at least four morphologically very distinct species of *Caulobothrium* beyond the two described species *C. opisthorchis* and *C. tetrascaphium*. *Urobatis tumbesensis* was found to host at least two species. Unlike *Caulobothrium* species from all other localities, the species parasitizing the two host species from the eastern Pacific Ocean grouped together in a subclade in the tree resulting from the phylogenetic analysis. Furthermore, species from the two hosts were intermingled across the tree topology. This is interesting given that *U. tumbesensis* occurs from Colombia to Peru and although *M. californica* is generally considered to occur only from Oregon to Baja California, there is some evidence it may also occur in the Galapagos Islands off Ecuador. The addition of sequence data for plerocercoids collected from several species of clams off California suggests that one or more of these species use clams as their final intermediate hosts.

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More than just a ball with a skirt: Advancing *Balanobothrium* and its relatives to a new order.

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Among the genera of tapeworms that parasitize orectolobiform sharks, *Balanobothrium* is perhaps the most puzzling. Only three of the 15 described species are currently considered valid owing to the superficial nature of the original descriptions and confusion surrounding host identities of the 12 other species. The three valid species all parasitize the zebra shark, *Stegostoma fasciatum*, off the coasts of India or Sulawesi. New material of *Balanobothrium* was collected from individuals of *S. fasciatum* off Australia, Malaysian Borneo, Tanzania, and Thailand. Specimens were examined with light and scanning electron microscopy and sequence data for the 28S rDNA (D1–D3) gene were generated. Somewhat unexpectedly, none of these specimens appear to be conspecific with any of the described species. In fact, this material appears to include at least 7 new species of *Balanobothrium*. Furthermore, the new species are regionally distributed across the distribution of the zebra shark. Three of the new species are from Malaysian Borneo, two are from Australia, two are from Tanzania, and one is from Thailand. Although all of these species exhibit the characteristic globose scolex consisting of four bothridia that are fused anteriorly and free posteriorly, each with a pair of small bipronged hooks that is typical of the genus, the worms range in size from a few millimeters to over 20 centimeters and bear proglottids with anatomies that are markedly different. Sequence data confirmed the distinct nature of the three species for which material appropriate for molecular work was available. The results of a Maximum Likelihood phylogenetic analysis, which included representatives of the closely allied genera *Pedibothrium* and *Pachybothrium*, confirm the mutual monophyly, and thus validity, of *Balanobothrium*, relative to these genera. The discovery of Southwell's type material of *Balanobothrium parvum* indicates that not only is the type host of this species *S. fasciatum* (rather than *Trygon* sp.), but also this species should be considered valid. This brings the total number of species in *Balanobothrium* to 11, making it the most speciose genus of elasmobranch-hosted cestodes. With a total of 50 species, the Balanobothriidae, including *Yorkeria* and *Spiniloculus*, is poised for erection to ordinal level.

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Persistence prevails: Do any genera of carcharhiniform sharks NOT host *Anthobothrium*?

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The genus *Anthobothrium* is one of the most poorly known genera of tapeworms parasitizing sharks. Its members are unusual among elasmobranch tapeworms in that their bothridia lack an apical sucker and instead bear one or two circular bands of musculature on their distal surfaces. Historically, molecular work has failed to definitively resolve the phylogenetic relationships of this genus; it is currently one of nine groups in the non-monophyletic order "Tetraphyllidea". Its members primarily parasitize carcharinid sharks, with 10 of the valid species described from the genera *Carcharhinus*, *Galeorhinus*, *Prionace*, and *Rhizoprionodon*. The eleventh species was described from the butterfly ray *Gymnura altavela*. These hosts were reported from Argentina, Australia, India, Iran, Tunisia, and the United States. The primary goal of this study was to examine additional genera and species of carcharhiniform sharks to expand our understanding of the host associations of *Anthobothrium*. The secondary goal was to further explore phylogenetic relationships of the genus. A total of 22 shark species in the following genera were examined for *Anthobothrium*: *Carcharhinus*, *Eusphyra*, *Glyphis*, *Hemitriakis*, *Laminopsis*, *Mustelus*, *Negaprion*, *Scoliodon*, *Sphyrna*, and *Triaenodon*. Also examined was a specimen of the butterfly ray *Gymnura natalensis*. These elasmobranchs were collected from Australia, Borneo, Korea, Mexico, Senegal, Solomon Islands, South Africa, United Kingdom, and the United States. A subset of worms found was examined with light and scanning electron microscopy and sequence data were generated for the D1–D3 region of the 28S rDNA gene. All species of elasmobranchs were parasitized by one or two species of *Anthobothrium*. In total, a minimum of 30 species were found, all of which appear to be new to science. Most species exhibited extremely low intensities of infection. Descriptive work focused on *Anthobothrium* parasitizing the second species of butterfly ray and the pairs of species parasitizing *Negaprion acutidens* and *Triaenodon obesus*. The tree resulting from a Maximum Likelihood phylogenetic analysis provides little evidence of host signal; species of *Anthobothrium* parasitizing different host genera, and even families, were found to be intermingled across the topology of the tree. However, evidence for establishment of this taxon at the ordinal level is mounting.

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Endoparasite diversity of subterranean rodents (Rodentia: Mammalia) and biogeographical hypotheses of the genus *Arostrilepis* Mas-Coma & Tenora, 1997 (Hymenolepididae: Cyclophyllidea), tapeworms from the Holarctic.

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Subterranean rodents are adapted to live underground with minimal surface dependence. Various species occur across four major zoogeographic regions: Ethiopian, Palearctic/Oriental, Nearctic, and Neotropical. A total, 282 different host parasite associations were detected using network analysis of the hosts and parasites. Six species of subterranean rodents harbor three tapeworm species of the genus *Arostrilepis* Mas-Coma and Tenora 1997 from the Holarctic region. Currently, there are 16 species of *Arostrilepis* occurring in 28 species of suitable hosts and have been reported from subterranean rodents from a southernmost record of about 36°N latitude to 75°N. Over 70% of the *Arostrilepis* host species are arvicoline rodents. The evolution and dispersal of these parasites remained unclear until the present time. However, the determination of the biogeographical origin of *Arostrilepis* is still controversial due to the quality and availability of its data. Here we present the comparison of two different methods with two datasets that showed opposing hypotheses about the biogeographical origin of these parasites. The resolution of this inconsistency will only be resolved when data are complete. It means that we need to collect more data.

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Molecular characterization of *Trichuris* spp. (Nematoda: Trichuridae) from rodents and marsupials from the Nearctic and Neotropical regions.

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Trichuris is a cosmopolitan genus with approximately 110 species of mammalian-dwelling parasites. In the New World, 30 of these species infect rodents of ten different families, including Caviidae, Cricetidae, Ctenomyidae, Dasyproctidae, Echimyidae, Geomyidae, Heteromyidae, Myocastoridae, Octodontidae, and Sciuridae. The diversity of the group lacks a phylogenetic context. Herein we present a phylogenetic analysis of 16 species, including ten collected from five families, including Cricetidae, Ctenomyidae, Cuniculidae Didelphidae, and Heteromyidae. The hosts and their parasites were collected in the United States, Mexico, Peru, and Argentina. Phylogenetic reconstruction based on the analysis of nuclear ribosomal genes (28S, 18S, ITS1-5.8S-ITS2) and newly sequenced partial mitochondrial genomes, generated through Illumina Sequencing, reveals at least four different lineages present in rodents from the New World. Each of these lineages is associated with Heteromyids (Mexico), Cricetidae: Sigmodontinae (Argentina), Cricetidae: Arvicolinae (Palearctic), and Cuniculidae/Ctenomyidae (Peru and Argentina), furthermore, there is one single lineage associated with marsupials. The analysis allowed us to establish the Mesoamerican distribution for parasites of *Heteromys gaumeri* and identify a new species present in *H. irroratus*. Further taxon sampling is necessary to assess the divergence times of these lineages and their macroevolutionary history across North and South America.

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A longitudinal snail-trematode survey at reclaimed wetlands in central Alberta.

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Every animal in an aquatic ecosystem can be parasitized by a digenean trematode. The ubiquity of digenean trematodes allows them to be employed as biodiversity indicator organisms due to their connections to other species in the environment. Even if their host is no longer present in the area, trematodes can act as a record of their presence. For the last decade, the trematode community of Alberta, Canada has been extensively studied resulting in the identification of 79 species. It is one of the most studied trematode communities in the world, making it ideal to study the relationship between biodiversity and disease, as well as the impact of biodiversity on host specialist and host generalist trematodes. Throughout the last four years, we employed trematode/snail sampling and traditional biodiversity survey methods to characterize the host-parasite communities at eight reclaimed wetland sites that differ in age but share the same geographic area. Snail collections occurred biweekly from June to September. Cercariae emerging from the collected snails were identified using DNA barcoding. Traditional biodiversity monitoring tools included benthic kick-netting, water sampling, field cameras and birdsong recorders. 1 979 of 22 396 snails were found to be infected with a digenean trematode (8.84%). Fifty-six species of trematodes from nine families have been identified at these sites, including 12 species that were not previously identified from Alberta, bringing the province-wide total to 91 species. We have also identified 96 species of vertebrate potential hosts and 59 species of potential invertebrate hosts. By combining the database of known trematodes in Alberta with the trematode and host data collected during this study, we can examine the relationship between host biodiversity and trematode species richness on a large scale and fill gaps in trematode lifecycles.

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Amphibian myiasis in North America: An update on host use, distribution and taxonomy of Calliphorids that infect amphibians.

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Currently, four families of dipterans have been implicated in amphibian myiasis across the world, including Calliphoridae, Chloropidae, Phoridae, and Sarcophagidae, some of which can cause considerable mortality in their amphibian hosts. Until recently, in North America, most reports (10/12) of anuran myiasis were attributed to two species of blowflies (Calliphoridae) including *Lucilia silvarum* (Meigen) and *Lucilia elongata* (Shannon). The latter species is exceedingly rare and its life history is unknown, but *L. silvarum* is common and was thought to be capable of being either parasitic or saprophytic in North America. In contrast, *Lucilia bufonivora* (Moniez) was thought to be an obligate parasite of amphibians and strictly distributed in the Palearctic. However, a morphological study in 2014 demonstrated that this species is established throughout southern Canada. More recently, a number of morphological and molecular studies have argued that *L. bufonivora* is also present throughout North America, and *L. silvarum* is now considered saprophagous in the Nearctic like they are in the Palearctic region. Here, I review our current understanding of the distribution of these flies and current problems with the taxonomy of these dipterans, including differences in host use and pattern of myiasis between European and North American specimens.

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A novel turtle disease? Histopathology of turtles infected with acanthocephalans.

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Acanthocephalans, known as thorny-headed worms, are a phylum of parasitic worms that infect the intestines of vertebrates as adults and have been reported to cause physical damage upon attachment to the intestinal wall with their proboscis. Although turtles commonly harbor hundreds of worms of the genus *Neoechinorhynchus* in their intestines, it is currently unknown if acanthocephalans cause any pathology to their turtle hosts. The goal of this study was to evaluate and characterize the degree of cellular pathology of multiple species of *Neoechinorhynchus* in turtle hosts. Red-eared slider turtles (*Trachemys scripta elegans*) were surveyed for helminth parasites in Oklahoma, U.S.A. Intestinal tissue samples with attached acanthocephalans were prepared for histological analysis, stained with hematoxylin and eosin and periodic acid-Schiff, and visualized under light microscopy. Additionally, non-infected turtle tissue was compared as a control. Pathological examination revealed mechanical damage to the intestinal wall of infected turtles, as well as signs characteristic of mucoid enteritis disease. These results suggest long-term anchoring of acanthocephalans and chronic host immune response. Depending on severity of infection, physiological consequences such as reduced digestion and absorption may impact the health of turtle hosts. Further investigation of turtle-acanthocephalan infections is necessary to better understand the implications of this disease.

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Niche expansion of malaria parasites in Caribbean lizards.

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There are 200 species of *Plasmodium* that use lizards as their vertebrate hosts. Despite their diversity in numerous traits, however, these malaria parasites remain understudied because they exist only in wildlife systems and do not directly impact human health. Genomic studies of bird-infective taxa identified a set of genes shared with the human malaria parasites, thus directly informing future translational research and highlighting the essentiality of these studies. Studying the genetic diversity of lizard malaria parasites promises to further inform future drug discovery studies by identifying additional conserved and essential parasite mechanisms. Here, we are beginning to unravel these mechanisms by studying hemoglobin digestion, an essential metabolic process, in *Plasmodium floridense*, *Plasmodium azurophilum*, and *Plasmodium leucocytica*. These species infect Anolis lizards throughout the Caribbean, and a single lizard host can be infected with one, two, or even all three of these parasites. In their vertebrate host, malaria parasites inhabit erythrocytes where they harvest the host's abundant supply of hemoglobin as a nutrient source. One noteworthy byproduct of hemoglobin digestion is free heme, which the parasites detoxify through hemozoin formation. Hemozoin is a hallmark of *Plasmodium* infection and has been implicated in the host immune response. However, two of the Anolis-infecting parasites do not produce hemozoin and have seemingly evolved alternative mechanisms to hemoglobin digestion. We hypothesize that this niche expansion is in response to coinfection, and the lack of hemozoin formation has allowed these two parasites to remain undetected by the lizard host immune system. To assess this hypothesis, we sequenced and assembled transcriptomes from fifteen infected and uninfected Anolis blood samples to obtain both parasite and

host information. Because there is no whole genome sequence for any lizard malaria parasite, this is the first sequence data available for these species. We have analyzed the transcriptomes for transcripts involved in the hemoglobin digestion pathway in the lizard malaria parasites as well as the host immune response to single and multi-parasite infection. Continued analysis of this host-parasite system will provide insights on trait evolution across the *Plasmodium* tree in the context of long-term evolution.

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Strongyle nemabiomes in horses: Prevalence, anthelmintic resistance, and factors affecting abundance.

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Small strongyles, also known as Cyathostominae, are common parasitic nematodes found in grazing horses and considered the most widespread helminth parasites affecting horses worldwide. There are over 50 different species and 14 genera of small strongyles that have been identified. They can cause larval cyathostominosis, a potentially fatal disease that occurs when large numbers of inhibited larvae are released from the intestinal mucosa. Horses with high numbers of small strongyles can also experience weight loss and unthrift, especially if they have underlying health conditions. To prevent the harmful effects of small strongyles, horses are regularly treated with anthelmintic drugs. However, the excessive use of these drugs has led to anthelmintic resistance. Resistance has been observed for all anthelmintic drug classes. Currently, ivermectin and pyrantel are the most used drugs used in horses. To examine the prevalence of small strongyles, as well as signs of anthelmintic resistance and factors that affect the occurrence and abundance, we used an internet survey distributed to farms and collected fecal samples before and after anthelmintic treatment. Thirty-two Swedish stables participated in the project, 16 with 135 horses treated with ivermectin and the other 16 with 132 horses treated with Pyrantel. The questionnaire responses were statistically analyzed, and fecal egg count (FEC) was performed. Larval cultures were set up, and DNA was subsequently extracted. The internal transcribed spacer 2 (ITS2) region was amplified using universal primers before NGS metabarcoding of the strongyle community (nemabiome). With a machine learning model, we found two factors affecting occurrence and abundance: horse age (for pyrantel, $p<0.05$) and FEC before treatment (for ivermectin, $p=<0.05$). We identified 32 species, with individual horses hosting up to 17 species in their nemabiome. The most frequently found small strongyle species were *Cyathostomum catinatum* and *Cylicocyclus nassatus*, making up more than half of the fractional abundance. Even though FEC decreased significantly after treatment, it did not change species diversity or alpha diversity. We conclude that selective anthelmintic treatment is effective in reducing the risk of anthelmintic resistance development, and we found no evidence of reduced efficacy in Ivermectin and Pyrantel.

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The prevalence of gastrointestinal parasites in breeds of cattle, goat and sheep in northern Pakistan.

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Globally, gastrointestinal (GI) parasites are a serious issue in livestock species important for food and agriculture. This study was conducted to assess the prevalence of gastrointestinal parasites in cattle, sheep and goat in District Lower Dir, Khyber Pakhtunkhwa, Pakistan. A total of 600 fecal samples were randomly collected including 300 from cattle and 150 each from sheep and goat for this study. The fecal samples were tested through direct microscopic examination, fecal flotation and sedimentation technique for the detection of GI parasites. The data was analyzed through Chi-square test using Statistical Package for Social Sciences (SPSS) to find out the effects of breed, age, parity, feeding practices, deworming status and herd size on the prevalence of GI tract in these species. The parasites were more prevalent (75.68%) in Jersey cross breed. Endoparasitic infection was higher (88.23) in male cattle. Greater than 2 years cattle had higher (71.02%) incidence of GI parasite. In herds of ≤ 5 animals had 67.28% positive samples and 6-8 animals per herd had 67.47% positive samples. *Toxocara vitulorum* prevalence was high (46%) in single parasitic infestation, while *Toxocara vitulorum* + *Haemonchus contortus* were more (34.6%) prevalent in double parasitic infection. *Haemonchus* + *Toxocara* + *Fasciola* spp. were detected more (33.3%) in triple parasitic infection in cattle. Among goat, Teady hairy goat and Beetal had 83.33 and 80% prevalence rate of GI parasite. Goats with 1-2 years had higher 81.53% prevalence rate. In herds of 16-20 goats 85.71% prevalence rate was reported. *Haemonchus* parasite was more prevalent in goat. Local breed of sheep had more (73.52%) prevalence rate of parasitic infection. *Haemonchus* parasite was more prevalent in sheep. It can be concluded from this study that all these livestock species in Lower Dir had highest prevalence of gastrointestinal parasites and need effective control measures to enhance productivity.

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First parasitological survey of bats (Mammalia: Chiroptera) from the Amazon River in Colombia.

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Bats (Mammalia: Chiroptera) comprise the most species-rich mammalian orders, with the highest diversity being located in the neotropics. Parasitological surveys of South American bats are scarce and no record of a bat parasite exists from the Amazon River within Colombia. The aim of this study is to describe a collection of bat parasites from the Amazon River at Leticia, Amazonas, Colombia. A total of 44 bats of 26 species, collectively exhibiting 4 discrete feeding habits were captured with a mist net during July through August 2021 and necropsied using standard parasitological methods. Most (26 of 44; 59%) bats were infected with at least one parasite; 15 were infected with both endoparasites and ectoparasites, 5 were infected with endoparasites only, and 6 were infected by ectoparasites only. The identified ectoparasites belonged to 18 species of 3 families (Hippoboscidae, Spinturnicidae, and Argasidae). The helminths identified belong to 3 phyla and 8 families (Nematoda: Capillaridae, Molineidae, Onchocercidae, Rictulariidae; Acanthocephala: Oligocanthorhynchidae; Platyhelminthes: Dicrocoeliidae, Lecithodendriidae, Hymenolepididae). A total of 5 parasites were identified to family, 12 to genus, 1 to subgenus, and 23 to species. Omnivorous bats had the highest diversity of parasites. All of these parasites comprise new records for the Amazon River within Colombia, and numerous new host records are presented.

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Influence of landscape heterogeneity on entomological and parasitological indices of malaria in Kisumu, Western Kenya.

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Background: Identification and characterization of larval habitats, documentation of *Anopheles* spp. Composition and abundance, and *Plasmodium* spp. infection burden are critical components of integrated vector management. The present study aimed to investigate the effect of landscape heterogeneity on entomological and parasitological indices of malaria in western Kenya. Methods: A cross-sectional entomological and parasitological survey was conducted along an altitudinal transect in three eco-epidemiological zones: lakeshore along the lakeside, hillside, and highland plateau during the wet and dry seasons in 2020 in Kisumu County, Kenya. Larval habitats for *Anopheles* mosquitoes were identified and characterized. Adult mosquitoes were sampled using pyrethrum spray catches (PSC). Finger prick blood samples were taken from residents and examined for malaria parasites by real-time PCR (RT-PCR). Results: Increased risk of *Plasmodium falciparum* infection was associated with residency in the lakeshore zone, school-age children, rainy season, and no ITNs ($\chi2= 41.201$, df= 9, $P < 0.0001$). Similarly, lakeshore zone and the rainy season significantly increased *Anopheles* spp. abundance. However, house structures such as wall type and whether the eave spaces were closed or open, as well as the use of ITNs, did not affect *Anopheles* spp. densities in the homes ($\chi2= 38.695$, df= 7, $P < 0.0001$). *Anopheles funestus* (41.8%) and *An. arabiensis* (29.1%) were the most abundant vectors in all zones. Sporozoite prevalence was 5.6% and 3.2% in the two species respectively. The lakeshore zone had the highest sporozoite prevalence (4.4%, 7/160) and inoculation rates (135.2 infective ites/person/year). High larval densities were significantly associated with lakeshore zone and hillside zones, animal hoof prints and tire truck larval habitats, wetland and pasture land, and the wet season. The larval habitat types differed significantly across the landscape zones and seasonality ($\chi2= 1453.044$, df= 298, $P < 0.0001$). Conclusion: The empirical evidence on the impact of landscape heterogeneity and seasonality on vector densities, parasite transmission, and Plasmodium infections in humans emphasizes the importance of tailoring specific adaptive environmental management interventions to specific landscape attributes to have a significant impact on transmission reduction.

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The toxicity of photosensitive insecticides against mosquitoes is bidirectionally modulated by the abundance of larval food.

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Insecticides are critical for controlling mosquito populations and mitigating the spread of vector–borne disease, but their overuse has led to the emergence of resistant populations. A promising alternative to classical chemical insecticides is photosensitive insecticides—or PSIs—which, when ingested and activated by light, generate broadly toxic reactive oxygen species (ROS) that kill mosquito larva. Here, we tested whether the PSIs, methylene blue and rose bengal, are a viable insecticidal strategy across the mosquito lineage and whether the toxicity of these PSIs varies in the presence of larval food. We found that methylene blue and rose bengal are phototoxic to both *Aedes aegypti* and *Anopheles gambiae*, but that methylene blue is over ten times more toxic than rose bengal. Although food stimulates PSI ingestion, we found that the addition of food has a bidirectional and dose-dependent effect on PSI toxicity: food decreases toxicity at lower PSI doses but increases toxicity at higher PSI doses. We predict that at lower PSI doses, most PSI-produced ROS are sequestered by the food, decreasing toxicity. However, at higher PSI doses, the increase in feeding results in an abundance of both sequestered and free PSIs which react with larval tissues and increase toxicity. Altogether, we reveal that (i) methylene blue and rose bengal are viable larvicides in evolutionarily distant mosquito species, (ii) methylene blue is more toxic than rose bengal, and (iii) the tandem application of larval food and PSIs can optimize larvicidal efficacy.

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History of parasite infection shapes the expression and magnitude of non-consumptive effects in cactophilic flies.

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Parasites can have non-consumptive effects (NCEs) in potential hosts, including changes in behaviour, morphology, and physiology even in the absence of infection. This ecology of fear is well-exemplified in a fly-mite system, *Drosophila nigrospiracula* and its parasite, *Macrocheles subbadius*. When chronically exposed to their ectoparasite, fly fitness is decreased even with no direct contact or possibility of infection. A previous experiment testing behavioural trade-offs between feeding and grooming found a strong interaction between previous and current parasite exposure: flies that were pre-exposed to mites (but never infected) showed increased feeding in the presence of mites, while naïve flies decreased their feeding, suggesting terminal reproductive investment or habituation. Here, I investigate how previous infection modulates fly behaviour in response to parasites. I hypothesized that previously infected flies will invest more energy in parasite avoidance (grooming) and evasive behaviours (jumping and tarsal flicking) in the presence of mites to avoid re-infection. I conducted 2x2 factorial experiments, where previous infection and current parasite exposure differed. Flies were first infected with mites, then the mites were removed after successful attachment. Control flies were similarly manipulated sans mites. Afterwards, evasive behaviours (jumping and tarsal flicking), parasite-avoidance (grooming), resting, and feeding were measured in a petri dish arena with or without mites. Having experienced the direct effects of parasitism, previously infected flies, compared to naïve flies, have increased their evasive and parasite-avoidance behaviours in the presence of mites, at the expense of resting and feeding, suggesting that more time and energy are allocated to avoiding new infections. Grooming, tarsal flicking, and jumping are energetically costly behaviours that divert time and energy away from life sustaining behaviours such as feeding. Changes in time allocation can have long lasting effects on individual fitness and population dynamics. The ecology of fear is far-reaching and can potentially affect hosts in ways previously unrecognized. In efforts to mitigate risk of parasitism, individuals can reduce host feeding and in turn reproductive success, leading to a decrease in population size.

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Mosquito ecology in zoological parks: Implications for zoonoses of mosquito-borne diseases.

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Zoological settings present unique opportunities to study mosquito ecology and to detect emerging diseases due to their diverse collection of captive animals that are potential blood-meal hosts. Mosquito-borne diseases (MBDs) are threats to the safety of those animals and the human population zoos attract. Mosquito-borne diseases have been responsible for the death of many captive mammals. Some MBDs like, avian malaria, have killed entire colonies of captive penguins. Animals in zoological parks can also serve as sentinels for MBDs capable of infecting humans. Despite this, there are only six zoos in the U.S. that have collaborated with scientists to establish mosquito surveillance systems, and only one of those locations have included epidemiological studies of parasitic diseases. Therefore, I initiated the first zoological mosquito surveillance study in Texas comparing mosquito distribution and abundance, as well as host blood-meal choice of mosquitoes at two zoos; the San Antonio Zoo and the Cameron Park Zoo in Waco, TX. Here I present preliminary results on mosquito species distribution and

abundance from the San Antonio Zoo during the summer of 2022. Two types of mosquito traps, BG-Sentinel® and Gravid traps, were set weekly at 11 sites from epi week 29 to 43 and mosquitoes were identified to species then stored in -80°C for future blood-meal analysis. *Culex quinquefasciatus* made up 82% of mosquitoes collected, followed by *Aedes albopictus* with 14%, and *Aedes aegypti* with 4%. We found that species diversity in the zoo was similar to that of the surrounding residential areas of San Antonio, which indicates a potential for zoonoses both to and from captive animals and the human populations. Partnerships between vector biologists and zoological parks are important to preserving the quality of life and healthcare of captive zoo animals and to improving emerging vector-borne disease detection and prevention.

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Parasites and the interconnected biosphere.

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In the current era, many students of biology first discover parasites and parasitism via the initial study of the ecology, behavior, or systematics of a species of free-living organism. While studying a free-living animal or plant (pick your favorite species), one discovers that there may be several species of parasites occurring in or on (or likely both) their study animals. After conversion, true parasitologists—those who are intrigued with the parasite's biology and intimate associations—may become intensely focused on a single group, like tapeworms of rodents or gregarines of beetles. Other students of parasitology may focus on the complete endoparasite fauna of a group of insects, fish, mammals, birds, amphibians, or reptiles. Work now being done in the HWML includes species discovery combined with ecological niche modeling, network analysis, and phylogenetic analyses to understand biogeographic distributions of parasites and hosts. Our recent work has shown that more than 60% of the 163 species of subterranean rodents globally are parasite data deficient. Meaning that no parasites have yet been recorded and published from these species. This is clearly a race against time as humanity is directly causing the loss of more species weekly than we are able to document and describe. Our current work indicates that species discovery and documentation are still at an early stage and must continue at pace.

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Parasites with agency: diversity and distribution of monoxenous nematodes in New World.

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Parasite diversity results from the historical interactions among the parasites and the compatible organisms that act as their hosts. These interactions are determined by the fluctuations of abundance resulting from geological and ecological changes. In the literature, scientists have documented the diversification of several parasite lineages, these appear to be disparate. Certain lineages of parasites matrilineally transmitted appear to codiverge with their hosts, in contrast rampant host switching prevails in lineages of parasites that require vectors. Yet, the phylogenies of parasites with direct patterns of transmission, including Aspidoderidae and Trichostrongylidae feature disparate patterns, perhaps because the fecal-oral route of transmission does not restrict exposure of the eggs to compatible sympatric hosts. As such members of the Aspidoderidae occur in mammals of different lineages that converge in semifossorial habits. Members of Trichostrongylidae are involved in host-switching events that appear to reflect the taxon pulse dynamics of six different lineages of rodents in the New World. I will use the phylogenies of these groups to show the different rates of parasite diversification.

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Characterization of immune-related genes in the invasive gekkonid species, *Hemidactylus turcicus*.

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Being the only ectothermic amniote clade, reptiles can serve as an effective evolutionary "bridge" in our understanding between the basal fish immune system and the mammalian system. Yet, studies on reptile immune defense in relation to helminth infections are virtually non-existent. As such, the development of resources in a reptile system amenable to study host-helminth interactions is necessary. To this end, we are developing genomic and transcriptomic resources for the invasive Mediterranean house gecko (*Hemidactylus turcicus*) because this reptile presents opportunities in studying vertebrate-parasite coevolution. First, the Mediterranean gecko is infected with several helminth parasites, some of which have high prevalence. Thus, the system has potential as a field-based model to study the evolution of vertebrate immune defense and helminth counter-defense. Second, because helminth infections in the gecko vary across locations in easily accessible locations (e.g., human habitation) across the southern U.S., the system can be used to test the role of parasite-mediated selection and spatial variation on the evolution of allelic variation at immune-related genes such as the major histocompatibility region (MHC). Currently, little is known about the MHC among gecko species. Here, we report on a draft assembly of *H. turcicus*. Using short and long-read data (i.e., Illumina and Nanopore reads, respectively), we generated a high-quality reference genome. We then searched this assembly to locate potential loci associated with the MHC region. Future work will utilize newer genomics tools (e.g., Hi-C) to fully characterize the MHC region. With such genomic resources at hand, we will be well poised to use the Mediterranean gecko as a model to address various aspects of vertebrate-helminth coevolution.

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Development of genomic resources for the gecko tapeworm, *Oochoristica javaensis.*

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Most genomic resources for parasitic platyhelminths are restricted to fish or mammalian host systems. The development of genomic resources for helminths that infect reptiles is necessary to provide a comparative basis for understanding the evolutionary dynamics of helminths across vertebrate taxa. We aim to develop genomic resources for a reptile-helminth system to fill this gap. Such a system can be further used to study the evolutionary interactions of vertebrate immunology and helminth counter-defense. To this end, we are developing genomic resources for the gecko tapeworm *Oochoristica javaensis*, a cyclophyllidean tapeworm native to Southeast Asia that uses gekkonid lizards as definitive hosts. We chose this tapeworm for several reasons. First, it is commonly found infecting the invasive Mediterranean gecko, *Hemidactylus turcicus*, in the southern U.S., and thus is amenable to field-based studies. Second, genomic resources are available for several mammalian cyclophyllideans of human or economic importance. Hence, *O. javaensis* can serve the role of a comparative base as a flatworm parasite of reptiles. We used short-read sequences, long-read sequences, and optical mapping data from several individuals to assemble a first draft genome of *O. javaensis*. The genome is 87.12 megabases long and has G + C content of 43.35%. Benchmarking Universal Single-Copy Orthologs

(BUSCO) scores were similar to or better than genome assemblies for other cyclophyllidean tapeworms (Eukarya, 66.8% complete; Metazoa, 85.9% complete). We identified 2 distinct mitochondrial haplotypes. Haplotype 1 is 13,885 base pairs and haplotype 2 is 13,914 base pairs long. As with other cyclophyllidean tapeworms, there are 12 protein coding genes, 22 transfer RNA genes, and 2 rRNA genes. The A + T content of haplotype 1 is 71.62% and haplotype 2 is 71.78%. Phylogenetic analysis of mitochondrial protein coding genes supports the placement of *O. javaenesis* in Cyclophyllidea. Future work in this system will address the evolutionary host-defense and parasite counter-defense interactions between the gecko host and its parasite. Moreover, we plan to characterize the tapeworm's gene expression across its developmental stages in its intermediate host (beetles) and definitive host (geckos).

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The transcriptome profiles of different life history stages of *Hematodinium perezi* (Dinoflagellata: Syndiniales) from the blue crab (*Callinectes sapidus*).

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The parasitic dinoflagellate *Hematodinium perezi* is an endoparasite in the hemolymph of marine crustaceans. All described syndinid parasites are considered parasitoids in the sense that they kill their hosts to complete their life cycles. The parasite has a complex life cycle with asexual division in several life history stages. Upon completion of development in the host, the parasite develops into either macro-, micro-, or pre-spore stages, which are released into the water column at extremely high densities by the dying host; however, each host only releases one type of dinospore. To date, sexuality in the order Syndiniales has rarely been documented and the roles of the different dinospores of *H. perezi* remain unknown. We collected dinospores released by individual crabs and extracted total RNA from four biological replicates from the three dinospore types as well as an asexual ameboid trophont stage from the hemolymph of the host. We sequenced the transcriptome using RNASeq. Each sample generated approximately 25 million reads. We anticipate finding differentially expressed genes in each life history stage that correspond with functional life history traits. Specifically, if the dinospores are analogous to gametes in Apicomplexan parasites, macro-dinospores or pre-spores (macrogametes or cysts) are expected to have up-regulated genes for cell development such as mitotic or meiotic progression and cyst wall formation; whereas micro-dinospores (if microgametes) are expected to have up-regulated genes for mobility such as those used in flagellar and microtubule synthesis. The asexual stage is expected to have up-regulated genes for within-host survival or metabolism such as surface proteins, enzymes used in protein digestion, signaling, and host cell regulation. Our findings on the transcriptomic profiles may provide insights into the cryptic sexual cycles of *H. perezi* and other members of their unusual parasitic order Syndiniales.

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Parasite assemblage characterization with a novel host signal reduction method and metabarcoding pipeline.

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Although current standard parasitological methods are of great utility, there remains a lack of high throughput alternatives to microscopy for large scale analysis of parasite assemblages. Whereas sequencing-based approaches (e.g., metabarcoding) of prokaryotes and fungi are commonplace, no single method for parasite metabarcoding has been adopted. We systematically assessed published parasite metabarcoding protocols and demonstrated issues with high off-target read abundance and primer complementarity. To address off-target signal, we designed a novel CRISPR-Cas9 based method that selectively digests vertebrate host DNA. When applied to blood and tissue samples, our method resulted in a mean 92 % decrease in host read abundance compared to no treatment, a 61 % decrease compared to the most commonly published blocking method, and allowed for detection of hemoparasite infections that would otherwise have been missed. To address primer complementarity, we designed new primers to recognize all parasites of vertebrate and used in silico PCR to show that only our new primers (n = 4) successfully amplified all parasite clades and had the highest overall taxonomic coverage as compared to published primer sets (n = 22). We then cloned 16 parasite DNAs to create a novel mock community standard spanning diverse parasite groups and used it to show that one of our new primer sets more closely recovered the underlying community than any other. When applied to clinical samples (n = 52) our new protocol (Vertebrate Eukaryotic endoSymbiont and Parasite Analysis, or VESPA) outperformed the “gold standard” method of microscopy with 51.3 % of identifications made by VESPA alone. VESPA identified taxa not found with microscopy, resolved a cryptic species complex not resolved by microscopy, and revealed greater prevalence and richness of parasitic organisms than microscopy.

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Assessing biodiversity in Madagascar with leech-derived iDNA: Methodological advancements and ecological insights.

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Global biodiversity is currently declining at unprecedented rates due to anthropogenic activities and interventions are needed to preserve remaining diversity. Baseline measures of biodiversity are needed against which to test the efficacy of such interventions in conserving species over time. Biodiversity inventories are used to establish these baselines, often relying on tools such as camera traps and Sherman traps, among others. With the advent of next generation sequencing, targeting residual species DNA from environmental sources (eDNA) has emerged as an efficient means of taking inventory of biodiversity. Of these environmental sources, blood feeding invertebrates have also been tested in their ability to preserve host DNA (invertebrate-derived DNA, iDNA). Here, I test and optimize the use of terrestrial blood feeding leeches of the family Haemadipsidae in sampling vertebrate biodiversity. I have developed a methodology for collecting leeches (*Chtonobdella* spp.), sequencing host DNA, and assigning taxonomic identity and I have implemented it in the forests of Madagascar, sampling some of the world’s most endangered biodiversity. I have shown that increasing barcoding loci increases taxonomic identifications, and that pooling individual blood meal DNA isolates for next generation sequencing is more efficient than individually targeting blood meals with traditional Sanger sequencing and fewer loci. I show that iDNA complements conventional biodiversity surveying methods, detecting

distinct vertebrate fauna. I have helped establish terrestrial leeches as valuable tools for biodiversity monitoring and I have improved our understanding of their behavior as understudied organisms.

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Isolating discrete life cycle stages of a cockroach-infecting Gregarine for gene expression analysis.

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Gregarines are apicomplexans that parasitize the intestinal epithelia of nearly all invertebrate clades via an oral-fecal pathway. Despite their abundant and diverse host-parasite relationships, Gregarines are largely understudied. Additionally, Gregarines closest relatives, Cryptosporidians, infect vertebrates and cause serious diarrheal disease in humans. Gregarines' similar lifestyle and life cycles to Cryptosporidians suggest that the largely understudied parasites may serve as a model for these human infectious parasites. *Blabericola migrator* is a species of Gregarine that solely infects Madagascar Hissing Cockroaches (*Gromphadorhina portentosa*). Gregarines have several distinct internal stages including: sporozoites, trophozoites (attached and detached), and those in the sexual stage of syzygy. In order to develop stage-specific Gregarine transcriptomes, we isolated three grossly defined subpopulations of internal stage Gregarines: attached trophozoites, detached trophozoites, and those in syzygy. Total RNA was isolated from four pooled biological replicates of each subpopulation and submitted for mRNA sequencing. Each subpopulation's transcriptome was compared to one another and to the published mixed internal stage's transcriptome to identify how gene expression is altered throughout the maturation of internal Gregarine parasites. Gene cluster analysis measured the variation among subpopulations and determined unique groupings for each subpopulation. The detached trophozoite subpopulation had the most abundance amongst its biological replicates most likely due to the biological variation of the subpopulation and its position as an intermediate between the attached trophozoite and syzygy subpopulations. Currently, gene ontology pathways are being analyzed. Once completed, these detailed findings will provide vital information regarding the stage-specific biology of the parasite and its interactions with its host.

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Surveillance for *Echinococcus multilocularis*, an emerging zoonotic parasite in New York wild canid populations.

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Echinococcus species are zoonotic cestodes that are of great medical and veterinary concern because they are known etiologic agents for alveolar echinococcosis and cystic echinococcosis in humans and wildlife. *Echinococcus* species are endemic to North America, inhabiting ranges that include the northern tundra and midwestern regions, with a historical absence in the northeast. Recent detections of *Echinococcus* spp. outside of their perceived range, in urban wildlife, and in humans represent a changing epidemiology of the parasite across North America. The introduction and spread of the more pathogenic and infective European variant of *Echinococcus multilocularis* may also be increasing the risk of human infection with the parasite, a hypothesis supported by the increase of alveolar echinococcosis cases in humans in Canada and the USA. We initiated a surveillance study for *Echinococcus* spp. in New York State following an incidental detection of *E. multilocularis* in a coyote in 2018, which represents one of the earliest detections of the parasite in the northeast. Between 2021-2023, 61 gastrointestinal tracts from coyotes (n=59) and foxes (n=2) harvested in New York were screened for *Echinococcus* spp. infection using a multiplex PCR and subsequent isolation of adult cestodes. We isolated *E. multilocularis* adults from five coyotes (8.2%) collected from three counties in the state. Analysis of the nad2 and cob mtDNA genes revealed that the New York parasites most closely grouped with parasite isolates from Europe, indicating a range expansion of the non-native variant of *E. multilocularis*. As definitive hosts, the vagile and commonplace nature of coyotes creates avenues for sylvatic transmission and spillover of the parasite to domestic animals and humans, and facilitates range expansion of the parasite in the USA. The emergence of *E. multilocularis* in the northeast USA presents a novel risk for wildlife, domestic animals, and humans, warranting further surveillance and research.

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The fruit fly, *Drosophila nigrospiracula*, exhibits threat-specific non-consumptive effects in response to parasite and predation risk.

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Non-consumptive effects (NCEs) collectively refer to changes in an organism’s behaviour, physiology or morphology that are caused by the perceived threat of predation or parasitism. Behaviours associated with NCEs have associated fitness costs as they can be energetically costly in and of themselves, decrease the time available for reproduction, or reduce access to high quality foraging grounds. NCE-related behaviours have frequently been investigated in predator-prey systems, but less often in parasite-host associations. Moreover, few studies have used the same species as both prey and host, i.e., to test the NCEs of exposure to either predator or prey in a species. We investigated the behaviour of the fruit fly *Drosophila nigrospiracula* exposed to visual and odor cues of an ectoparasitic mite (*Macrocheles subbadius*) or a predatory spider (*Salticus scenicus*). Using video analysis of fly behaviour, we first established that *D. nigrospiracula* could differentiate between parasitic and non-parasitic mites, and between predatory spiders and non-predator insects of a similar size. We then demonstrated that flies showed diametrically opposed responses to parasite vs. predator in certain behaviours; relative to controls, flies moved faster and spent more time grooming in the presence of mites but slower and less time grooming in the presence of spiders. These behaviours reflect a threat-specific response, as grooming and evasion are defence mechanisms against the attachment of comparatively slow moving ectoparasites such as mites, while slower movements and reduced extraneous activity such as grooming are appropriate defences against the spider, an ambush predator. These responses show that *D. nigrospiracula* possesses a degree of behavioural plasticity and a sophisticated ability to tailor their NCE-related behaviours to predator or parasite threats. Our results also suggest that there may not be a predictable, linear relationship between the magnitude of an NCE behaviour and the fitness cost of the threat-type, and so may provide new perspectives on how parasitism should be incorporated into NCE research.

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Downstream effects of changing life cycle complexity: testing the mating system and inbreeding depression among natural populations of the trematode *Alloglossidium progeneticum*.

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Evolutionary changes in parasite developmental and/or life cycle patterns can have subsequent consequences for ecological and evolutionary processes within parasite populations. Indeed, the mating system model of complex life cycle evolution by Brown et al. (2001) is based on the assumption that a shorter life cycle requiring fewer hosts is likely to result in more inbreeding, thus the model predicts that the evolution of a truncated life cycle is only possible if there is no resultant inbreeding depression. Several populations of the trematode *Alloglossidium progeneticum* have evolved a facultatively precocious life cycle, where sexual development can occur within the 2nd intermediate crayfish host. Here, the precocious life cycle likely leads to high levels of inbreeding as individuals are forced to self-mate while encysted in their crayfish hosts. Other populations of *A. progeneticum* have maintained the ancestral 3-host life cycle where sexual reproduction occurs among free adult stages in the intestine of catfish definitive hosts. Using microsatellite genetic markers to estimate the mating system, we aimed to confirm inbreeding in 5 precocious populations in Georgia compared to 4, obligate 3-host life cycle populations in Louisiana, Arkansas, or Texas. All populations were collected in 2018, and genetic estimates of selfing rates were derived using several methods: deviations from Hardy-Weinberg equilibrium (quantified via F_{IS}), identity disequilibrium, and model-based Bayesian estimators. Moreover, we provide a unique demographic estimator of the selfing rate based on parasite mean abundances in crayfish and catfish hosts and accounting for relative catfish to crayfish population sizes based on our sampling. By comparing the demographic and genetic selfing-rate estimates, we further test the mating system model prediction of no inbreeding depression for the abbreviated life cycles of precocious populations. The latter method provides a novel means to test inbreeding depression from field-based collections. While changes in life cycle patterns are in themselves fascinating evolutionary outcomes, our results highlight how such changes further impact evolutionary mechanisms operating within parasite populations.

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Patterns of asynchrony in helminth parasites of centrarchids in J. Strom Thurmond Lake, South Carolina.

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The likelihood of metapopulation persistence is dependent upon recolonization of local extinction events (the so-called “rescue effect”), which in turn is made more likely when the component subpopulations exhibit asynchronous subpopulation dynamics. The influence of stochastic environmental effects on subpopulation dynamics, combined with the tendency for these stochastic effects to be spatially correlated, leads to the prediction that the degree of asynchrony between subpopulations should covary positively with inter-site distance. Fishes within the genus *Lepomis* were collected monthly from 4 localities along the eastern shoreline of J. Strom Thurmond Lake, from July 2016 to July 2017. Parasite abundances were correlated among the localities in a pairwise fashion to generate metrics of subpopulation synchrony, and the degree of synchrony was examined for correlation with between-site distances, habitat characteristics, and centrarchid community structure.

Of the 7 parasite species examined, 2 (*Tylodelphys scheuringi* and *Crinicleidus* sp.) demonstrated correlations between asynchrony and distance, and 2 species (*Posthodiplostomum minimum* and *Spinitectus* sp.) had patterns of asynchrony that could not be associated with any of the site variables measured. Three parasite species (*Clavunculus bifurcatus, Crepidostomum cornutum*, and *Neoechinorhynchus cylindratus*) had patterns of asynchrony that covaried with habitat variables, which, in concert with the steady decline in the water level of the reservoir over the course of the investigation, suggests that large-scale effects can be "refracted" by differences in habitat characteristics, resulting in asynchrony over relatively short distances.

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Assessment of anthropogenic impacts on the aquatic environment and biodiversity in the Katangese Copperbelt Area (DR Congo): a parasitological approach.

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The field of fish parasitology, which was unexplored for the Upper Congo Basin until a few years ago, has seen an increased interest in the last decade in monogenean fish parasites. These monogeneans are being included in policy-relevant studies as potential bio-indicators of pollution. The Upper Congo Basin is an area with high mining potential and exploitation and has particular interest in biomonitoring. Monogeneans are sensitive to environmental changes, highly host specific, ubiquitous and have a direct lifecycle. Unfortunately, they are not well studied in many ecoregions of the Upper Congo Basin. This study aims to use monogeneans as bio-indicators of water pollution in the Katangese Copperbelt Area (KCA). As a starting point, we carry out an inventory of the parasitological fauna of the fish with the highest economic value and most abundant in fishermen's catch in two ecoregions spanning the KCA. Two fish families, Cichlidae, three fish species (*Oreochromis mweruensis*, *Coptodon rendalli* and *Serranochromis macrocephalus*) and Clariidae, four fish species (*Clarias ngamensis*, *C. gariepinus*, *C. buthupogon* and *C. stappersii*), were selected. Monogeneans were isolated from the gills and mounted on glass slides with ammonium picrate-glycerin for identification based on morphological analysis. Species richness was reported, and infection parameters of the retrieved parasites were calculated. A total of 13 parasite species were recorded on cichlid fishes, and ten monogenean species from clariids. The prevalence of monogeneans on cichlids ranged between 2 - 92%, while on clariids, it ranged between 7 - 67%. Mean intensities were between 1 - 17 ± 24 on cichlids, and 1 to 17± 21 on clariids. Based on preliminary results, monogenean species diversity and their respective epidemiological indices are low in sites near mining areas (source of pollution) and increase with increasing distance from them. This study highlights the potential biodiversity still to be explored in the Upper Congo Basin and it will serve as an important baseline for the assessment of water quality using bio-indicators in the KCA.

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Illumina sequence data and morphological variability lend support to further diversity among North American *Gyrinicola* (Nematoda: Oxyuroidea).

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Gyrinicola Yamaguti, 1938 are found within the intestinal tract of numerous larval anuran species in Europe, Asia, South America, and North America, with 6 valid species. Within North America, only *Gyrinicola batrachiensis* is recognized. This species, however, has a controversial taxonomic history; 2 species were once recognized in North America: *Pharyngodon batrachiensis* and *Pharyngodon armatus*, which were synonymized due to morphological and reproductive similarity. Previous analyses of nuclear ribosomal genes, using specimens from across the country, suggested cryptic diversity may be present in North America. To further investigate possible species diversity of *Gyrinicola*, an integrative study utilizing morphology and genetic information of specimens from a wide range within the United States was performed. Phylogenies of the nuclear ribosomal genes (*28S*,*18S*, *ITS1-5.8S-ITS2*) and newly sequenced partial mitochondrial genomes, generated through Illumina Sequencing, suggest at least 4 species groups exist among the nematodes presently sampled when evaluated with bPTP species delimitation methods. A Multiple Analysis of Variance (MANOVA) of female and male characteristics informed by the previous genetic investigation further supported the existence of 4 species groups. These results stress the importance of integrative investigations of oxyurid nematodes. This study represents the first genetic evaluation of the diversity of the genus in North America, the first comparison of these nematodes collected from such distant localities, and the first sequencing of mitochondrial genes for these nematodes. Furthermore, amplification of mitochondrial genes may allow for the production of new primers to aid in further investigation of these nematodes by future researchers.

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Supplemental description and phylogenetics of *Dracovermis occidentalis* (Digenea: Liolopidae) infecting the intestine of an American alligator, *Alligator mississippiensis* from the Mobile–Tensaw River Delta, Alabama, USA.

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As part of a parasitological survey of American alligators, *Alligator mississippiensis* (Crocodilia: Alligatoridae) in the southeastern United States, we examined several alligators from Louisiana, Alabama, and South Carolina. During the annual Alabama alligator hunt (August 2022), we collected several specimens of *Dracovermis occidentalis* (Platyhelminthes: Digenea: Liolopidae) from the intestine of a single alligator (alligators from Louisiana and South Carolina were not infected). These liolopid specimens were heat-killed and fixed in 10% formalin for morphology or preserved alive in 95% EtOH for DNA extraction. Liolopids comprise 5 genera of 15 species: *Liolope* spp. infecting giant salamanders, *Helicotrema* spp. infecting turtle and lizards, *Harmotrema* spp. infecting snakes, *Paraharmotrema* infecting turtles, and *Dracovermis* spp. infecting crododilians. Based on our newly-collected specimens, we emend *Dracovermis* and provide supplemental observations of *D. occidentalis*. Our specimens were assigned to *Dracovermis* because they lack tegumental spines/scales and have testes in the posterior 1/3 of the body, a pretesticular cirrus sac, a spined and eversible cirrus, a bipartite seminal vesicle, and a post-acetabular vitellarium. A phylogenetic analysis of the D1–D3 domains of the nuclear large subunit ribosomal DNA (28S), which is the largest taxon sampling for liolopids to date, recovered Liolopidae as monophyletic but did not support the previous assertion of parasite-host cophyly among liolopids and their vertebrate hosts. This is the first morphological study of new specimens of *Dracovermis* since the genus was proposed; the first record of a liolopid from Alabama; and the first phylogenetic analysis that includes *Dracovermis*.

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Hiding in plain sight: what can endogenous development and genetic sequencing tell us about coccidia diversity in lizards?

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Classification of coccidia at the generic and family levels was traditionally based on the life cycle, and the number of sporocysts and sporozoites in the exogenous stages (oocysts). For example, 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. To address this issue, we examined the coccidia 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 world-wide. Our 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.

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A new gregarine infecting the dune beetle, *Edrotes ventricosus*, (Coleoptera: Tenebrionidae) in the Mojave desert of California.

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Gregarines are common protozoan parasites of insects, particularly beetles. For 25 years, our lab has surveyed the gregarine fauna of tenebrionid beetles of the Great Plains and we have recently expanded our survey to the American West, particularly in desert biomes. In March 2019, we surveyed tenebrionid beetles for new gregarine parasites in the Mojave Desert north of Joshua Tree National Park in San Bernardino County, California. Beetles were hand collected using a headlamp to search the desert floor

for active beetles after sundown. Gregarine gametocysts were collected from frass after holding beetles overnight and then beetles were dissected and gregarine parasites were fixed on cover slips for subsequent staining and morphological analysis. Gametocysts were stored for oocyst collection or subsequent genomic analysis. Forty-eight of 57 (84%) dune beetles, *Edrotes ventricosus*, collected were infected with a previously unknown species of gregarine of the genus *Stylocephalus*. Species of *Stylocephalus* are differentiated by the morphology of the epimerite, overall size and morphology of the gamont, and the size and shape of the oocyst. Of these, overall gamont size and oocyst morphology are most useful in differentiating species. Two species of *Stylocephalus* are described from the New World, all parasitic in tenebrionid beetles. *Stylocephalus giganteus* is at least twice the size of the new species from dune beetles. The new species is notably larger than *Stylocephalus occidentalis*, with longer gamonts (575 vs 381 µm, respectively) and larger oocysts (11.3 x 8.3 vs 9.8 x 7.9 µm, respectively). Our morphological data demonstrate that the new species from *E. ventricosus* is distinct from known North American species of *Stylocephalus*. We are now sequencing the 18s, ITS, and 28s gene regions from the new species as part of a larger phylogenetic analysis of gregarines from North American Tenebrionids.

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Molecular phylogenetics of the ectoparasitic sucking louse genera *Hoplopleura* and *Polyplax* (Phthiraptera: Anoplura, Hoplopleuridae, and Polyplacidae).

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Sucking lice (Phthiraptera: Anoplura) are obligate and permanent hematophagous ectoparasites of placental mammals. Globally, more than 550 valid species of sucking lice have been recognized belonging to 50 genera and 15 families, where almost 60% of them are ectoparasites of rodents. Traditionally, morphology-based phylogenies have been used to describe the relationships among the groups inside Anoplura, however, a recent molecular phylogenetic analysis demonstrates that some clades of sucking lice are not monophyletic. The discrepancy between these two datasets indicates that further studies are needed to reevaluate the phylogenetic relationships across the group. Within Anoplura, the genera *Hoplopleura* and *Polyplax* are the most diverse genera of sucking lice, with 164 species and 82 species, respectively. Historically, these genera have undergone several taxonomic changes and their limits have been only tested in a morphological framework, neither their monophyly nor internal relationships have been analyzed in a molecular phylogenetic context. Consequently, the purpose of this project was to test the monophyly of the genera *Hoplopleura* and *Polyplax* using a molecular approach. We used four molecular markers (16s, 18s, COI, and EF-1α) broadly used in Hexapoda systematics and we placed our resulting hypotheses in the context of the previously reported phylogenies of anoplurans to determine the internal relationships and their position within the Anoplura as well as to analyze their host associations by incorporating the phylogenies of their hosts.

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Lamproglena cleopatra (Copepoda: Lernaeidae) from *Labeo victorianus* (Cypriniformes: Cyprinidae) with first ribosomal and mitochondrial genetic data.

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The family Lerneidae Cobbold, 1879 comprises among others the cosmopolitan parasitic freshwater copepods of the genus Lamproglena von Nordmann, 1832. This genus is regarded the oldest and second largest member of Lerneidae of which the females are parasitic on the gills of teleost fishes. To date 38 nominal taxa have been morphologically described with only four species genetically characterised using 18S and 28S ribosomal DNA (rDNA). This study was carried out in River Nyando of Lake Victoria Basin, Kenya, where a parasite of the genus *Lamproglena* was collected from gills of a cyprinid Ningu Labeo victorianus Boulenger, 1901. The parasite was studied morphologically using scanning electron microscopy (SEM) and molecular analysis and identified as *Lamproglena cleopatra* Humes, 1957. SEM analyses revealed slight additional taxonomic features on antennules, oral apparatus, leg 2-5 and caudal ramus which has not been previously recorded. Results of 18S, 28S rDNA and cytochrome c oxidase subunit 1 mitochondrial DNA dataset showed that the species was distinct from those of other *Lamproglena* taxa retrieved from GenBank but placed the species within this genus. This is the first DNA study of this parasitic copepod. The cox1 mtDNA data set was provided for the first time for this genus. Even though the results presented here greatly adds to the molecular data available for *Lamproglena*, there are still 34 (>89%) taxa for which no genetic data are available. The interpretation of the results presented here are thus preliminary and much more data are required before the phylogeny of this genus can be well studied. This work was supported in part by DSI-NRF SARChI Chair in Ecosystem Health (101054).

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Diversity of helminths in California birds in relation to host traits.

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The diversity of helminths found within different bird species has been the focus of many parasitological studies, but it still begs the question: why do intestinal helminth communities vary so much even within individuals of the same species of definitive host? In this project, we investigated the species richness, identity, and magnitude of infection of helminths in 17 different species of birds representing the orders Anseriformes; Pelecaniformes; Passeriformes Accipitriformes; Suliformes; and Galliformes from in and around the San Francisco Bay Area of California, USA. We compared these patterns to host traits with a focus on feeding behavior (carnivorous/herbivorous), migration behavior, and habitat location (freshwater/saltwater). Our second objective for this project involved determining the definitive host of the trematode, *Ribeiroia ondatrae*, in the birds from California. As the definitive host of *R. ondatrae* is still relatively unknown within this state, this project aimed to determine the primary definitive host species. A total of 56 gastrointestinal tracts were provided through several different local collaborators, such as wildlife rehabilitation centers and hunting groups. We examined the gastrointestinal tracts to identify and observe helminths following standard procedures. Helminths were identified morphologically to the lowest taxonomic level. Complementary genetic identifications are ongoing. Out of 56 birds from 13 counties in CA, 89% were infected by helminths; we identified 7 species of nematodes, 12 species of trematodes, 3 species of cestodes, and 5 species of acanthocephalans. We observed that certain helminths were found across multiple host species while others were found primarily in a single species. For example, we found the trematode *Clinostomum sp.* in five different bird species. Through the analysis of host traits, we observed a variation of helminth diversity and abundance in relation to specific host traits, such as their migratory behavior. By comparing the richness and composition of helminths to common host traits, we aim to further understand the relationship between specific bird host traits and their relation to the helminth diversity we observe.

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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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Parasites of freshwater mussels of Texas and the effect of trematode parthenitae on host reproduction and mortality.

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In Texas, 15 of ~52 freshwater mussel species are listed as state-threatened. Parasitism rates are not well characterized, but castrating trematode larvae have been reported in the gonads of *Cyclonaias petrina*, which is proposed for listing under the U.S. Endangered Species Act. Our research aims to investigate: (1) the parasites of mussels in Texas and factors related to infection; and (2) the effects of trematode larvae infection on reproduction. Collections occurred in central and east Texas on the San Saba River from April to November 2021 and on the Sabine, San Jacinto, and Brazos rivers in August 2021. Collected mussels were dissected, and their parasites were preserved and stained. To evaluate impacts on reproduction, we marked mussels at two sites on the San Saba River in July 2021. Recapture

occurred in November 2021, April 2022, and December 2022. All mussels were screened stream-side for trematode larvae. We used Generalized Linear Models to assess factors affecting the prevalence and abundance of each parasite. Adult aspidogasters were found in three mussel species, and abundance varied among sites. Mites were found in all but one mussel species, and abundance was positively related to host body condition, although likely affected by two large, heavily infected hosts. Of the collected mussels, six of nine species were infected with trematode parthenitae of Bucephalidae and Gorgoderidae, including 53.3% of *C. petrina*. Hosts infected with trematode parthenitae had no visible gametes. For the mark-recapture study, 1/1 *Potamilus fragilis*, 10/11 *C. petrina*, 5/10 *Cyclonaias pustulosa*, 1/4 *Utterbackia imbecillis*, and 2/32 *Tritogonia verrucosa* were infected with bucephalid sporocysts. One of the eighty recaptures exhibited infection loss, indicating possible recovery. The high infection rate of larval trematodes in the state-endangered *C. petrina* is troubling because impacts on reproduction combined with declining populations can erode long-term viability. The high mortality rate (7 out of 10 recaptured) of *C. petrina* is further worrisome. These deaths may be caused by endangered mussels being overburdened by infection, especially during environmental stressors. To better assess changes in infection and mortality in at-risk species, we will continue to recapture marked mussels at these sites.

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Parasites, pathogens, and the extended specimen.

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Parasites are of vital interest and concern in efforts to address global challenges on many fronts, from emerging pathogens and zoonotic pandemics to biodiversity loss and climate change. Museum collections of parasites and hosts provide critical infrastructure related to species occurrences, interactions, distributions, and temporal baselines that allow assessments of the extent and direction of environmental change. Physical voucher specimens also validate taxonomic changes, genomic data, and a variety of studies using new technologies that are sample based. Finally, specimens facilitate replication and extension of previous research, critical aspects of the scientific process. Museum specimen data digitized and made available online are now being integrated across collections and institutions and contribute to public datasets (GBIF, VertNet, GGBN, iDigBio, GloBI, TPT, SCAN) that are accessible for discovery, education, and research. The Museum of Southwestern Biology has focused on integrating parasite specimens and data with vertebrate host data for imperiled high latitude, aridland, and tropical ecosystems through the Rausch Helminthological Collections, Sevilleta LTER Rodent/Parasite Project, Global Schistosome Diversity, Beringian Coevolution Project, Panama Hantavirus Project, PICANTE, Steppe-NET, and Terrestrial Parasite Tracker (TPT) projects that have facilitated parasite digitization initiatives. Using the Arctos Collection Management System, we are developing tools to capture and manage host/parasite data and make these data publicly accessible and discoverable via the Arctos online search portal. These tools include url-based relationships between records to link hosts, parasites, associated images, media, genomic data and publications within and across collections and institutions; host and parasite observation portals for capture, linkage, and discovery of verbatim and unvouchered host/parasite specimen data; automated linkage of more complex parasite/host assemblages in a dashboard “entity” portal with shareable url; and most recently, documenting host examination/detection of parasites and pathogens to capture testing methods and parasite/pathogen prevalence data. We demonstrate these tools for managing and discovering parasite/host/pathogen data and discuss future community directions.

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Alteration of behavior in aquatic snails due to larval infections of *Paragordius varius.*

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Paragordius varius [Nematomorpha] is a freshwater species of horsehair worm with a complex lifecycle that entails using midge larvae as the paratenic host, which transfers the cyst stage from the aquatic to the terrestrial environment after metamorphizing into adult midges. These midges are then consumed by the cricket definitive host where *P. varius* matures into adults. Once mature, *P. varius* manipulates the behavior of the cricket, causing it to enter water where the fully developed worms emerge, mate, and lay eggs which hatch into larvae that encyst in midges, completing the life cycle. *P. varius* larvae can also encyst in aquatic snails, but it is unclear if snails can serve as a viable infection route to the definitive host. We hypothesize that if aquatic snails can serve as a paratenic host for *P. varius*, then infected snails will spend more time above the surface of the water, and thus be more likely to be consumed by a definitive host. Preliminary studies found evidence implying such an effect on the behavior of infected snails during the hours of 8am – 10am. In this study, we use distance above the waterline and activity of 7 experimentally infected and 9 sham-infected snails in a 2.5 hour period to determine if infected snails are manipulated by *P. varius*. We use EthoVision software to track snail movement in an aquarium. Our results found that uninfected snails spent significantly more time at the surface (36.9 minutes) than uninfected snails (16.3 minutes). Because such behavior provides no obvious benefit to either the parasite or the host, we hypothesize that this altered behavior is likely a by-product of the infection. If this is the case, then after dissecting the snails, we should see that snails with more cysts spend less time at the surface than snails with fewer cysts.

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Behavioural changes in potential host snails (*Stagnicola elodes*) depends on abiotic and biotic Factors.

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Host manipulation is often documented for parasites that use trophic transmission. However, many parasite life cycles require non-trophic transmission, where larval parasites must encounter and penetrate the next host in the life cycle. The role of host manipulation in non-trophic transmission is unclear despite its potential benefit to larval parasites because several abiotic (temperature, illumination) and biotic (heterospecific vs conspecific) factors may influence the outcome. In echinostome trematodes, non-trophic transmission commonly occurs between gastropod first hosts and a wide variety of invertebrate second hosts (including snails). In a lab-controlled environment, these parasites manipulate snail behaviour by altering the navigational behaviour of uninfected second hosts to infected first hosts. However, the generality of host manipulation is unknown because attraction has only been demonstrated in specific abiotic and biotic conditions (e.g., ambient lighting, room temperature water, and heterospecific stimuli). Increased water temperature and illumination affect the thermal biology of snails and larval trematodes, but it is unclear how these abiotic factors influence the navigational behaviour of snails and parasite-modified behaviour. We hypothesized that snails would become more active with higher illumination and water temperature, and that attraction between conspecifics would increase when one individual was parasitized. Using a Y-maze, we assessed the

behaviour of uninfected *Stagnicola elodes* in response to no stimuli (negative control), vegetation (positive control), uninfected conspecifics, and echinostome-infected conspecifics (n = 30 trials per treatment). To determine how abiotic conditions affected snail navigation, control treatments were conducted at two temperature and light conditions (mean 21.1°C and 165 Lux; mean 25.3°C and 5500 Lux). We video recorded the behaviour of each responder for 30 minutes to measure the average distance from the stimulus, the time spent in direct contact, the time spent in each arm, and the arm first entered. Results showed that higher illumination and temperature conditions increased the movement of the responder, which led us to conduct the remaining of the treatments using these conditions. We will discuss the results of conspecific attraction with and without parasitism. This research broadens our understanding of the context-dependent nature of parasite-modified behaviour both in lab and natural environments.

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Benchmarking Q20+ ONT MinION long-read sequence data for generating reference-quality genomes for parasitic nematodes.

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Parasitic nematodes are etiological agents of both acute and chronic disease in humans, livestock, and companion animals. Recently, the study and treatment of parasitic nematodes has been revolutionized by technological and methodological advances in genomics. Though remarkably impactful, these advances have also underscored a critical issue: Generating short-read DNA sequence data (and building and maintaining the infrastructure needed to do so) is expensive, making genomic resources largely inaccessible. The Oxford Nanopore Technologies (ONT) MinION is an inexpensive and portable next-generation sequencing platform capable of producing ultra-long DNA sequences ideal for whole genome assemblies; however, poor read level accuracy has hindered widespread adoption of the platform. Recent advances in ONT chemistries have produced more accurate and reliable read level data. To determine if whole genome assemblies using only ONT MinION data reflect current gold standard hybrid assemblies, we generated a complete genome from a single adult worm of the filarial nematode *Brugia malayi* using only ONT MinION Q20+ sequence data. We compared the ONT MinION-only assembly to a reference genome generated using a hybrid assembly approach, and assessed contiguity, completeness, and accuracy using QUAST, BUSCO, and MUMmer4, respectively. Ultimately, we generated an 88 gigabase assembly in 85 contigs that covers 99.8% of the reference genome. The two assemblies both contain >99% of BUSCOs known for the phylum Nematoda and are >99.9% identical when considering only single base pair mismatches. These results highlight the ONT MinION and its Q20+ platform update as an accessible stand-alone alternative to traditional sequencing approaches for the generation of whole genome assemblies for parasitic nematodes.

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An array of PCR-RFLP markers for differentiating between *Schistosoma haematobium* and *S. bovis* in field settings.

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Parasitic blood flukes in the genus *Schistosoma* are known to hybridize in the laboratory. Hybridization between S. haematobium, a human parasite, and S. bovis, a cattle parasite, is of particular concern due to its potential implications for zoonotic transmission and the complex strategies required to control both human and animal pathogens. Multiple genotyping studies have identified S. bovis mitochondrial DNA in miracidia carrying S. haematobium ribosomal DNA, leading to a widespread assumption that hybridization between these species occurs commonly. By comparison, studies employing a large number of autosomal markers, including whole-genome single nucleotide variants and microsatellites, have failed to identify early generation hybrids between S. haematobium and S. bovis, and instead find that these species are strongly differentiated. We aimed to develop a simple field applicable approach, utilizing Restriction fragment length polymorphisms (RFLPs), to distinguishing S. haematobium and S. bovis in the field. We analyzed whole-genome sequence data from 167 S. haematobium and S. bovis samples collected across Africa and reference genome of both species to design RFLP loci for species differentiation. We identified 269,779 single nucleotide variants distinguishing the two species and generated 34 candidates autosomal RFLP markers. These markers were further tested in the laboratory using a combination of field and lab-derived S. haematobium and S. bovis samples. Our specific intention is to use these markers to distinguish S. bovis and S. haematobium cercariae shed from Bulinus spp. snails collected in the field. More generally, we believe that genotyping these autosomal SNPs (using RFLP or more high throughput approaches) will allow more effective differentiation of S. haematobium and S. bovis compared to markers (COX, ITS, 18s) currently in use.

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A genome sequence for *Biomphalaria pfeifferi,* the major vector snail for the human-infecting parasite *Schistosoma mansoni.*

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Biomphalaria pfeifferi is the world's most widely distributed and commonly implicated vector snail species for the causative agent of human intestinal schistosomiasis, *Schistosoma mansoni*. In efforts to control *S. mansoni* transmission, chemotherapy alone has proven insufficient. New approaches to snail control offer a way forward, and possible genetic manipulations of snail vectors will require new tools. Towards this goal, we have sequenced the genome of a specimen of *B. pfeifferi* from Kenya. Based largely on PacBio High-Fidelity long reads, the genome assembly size is 772 Mb, smaller in size than known genomes of other planorbid schistosome vectors. In a total of 505 scaffolds (N50 = 3.2Mb), 430 were assigned to 18 large linkage groups inferred to represent the 18 known chromosomes, based on whole genome comparisons with *Biomphalaria glabrata*. The annotated *B. pfeifferi* genome reveals a divergence time of 3.01 million years with *B. glabrata*, a South American species believed to be similar to the progenitors of *B. pfeifferi* which undertook a trans-Atlantic colonization < five million years ago. The genome for this preferentially self-crossing species is less heterozygous than related species known to be preferential out-crossers; its smaller genome relative to congeners may similarly reflect its preference for selfing. Expansions of gene families with immune relevance are noted, including the *FReD* gene family which is far more similar in its composition to *B. glabrata* than to *Bulinus truncatus*, a vector for *Schistosoma haematobium*. Provision of this annotated genome will help better understand the dependencies of trematodes on snails, enable broader comparative insights regarding factors contributing to susceptibility/ resistance of snails to schistosome infections, and provide an invaluable resource with respect to identifying and manipulating snail genes as potential targets for more specific snail control programs. This study was supported by NIH grant award number R37AI101438.

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The journey of tsetse transmitted trypanosomes: from the skin invasion to a systemic infection.

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The African trypanosome species (*Trypanosoma brucei rhodesiense* and *T. b. gambiense*) are transmitted by tsetse flies (*Glossina* sp.). Due to major control efforts, the annual number of reported human cases has declined to about 800, with a roadmap to reach elimination of the gambiense form by 2030. However, the major challenges that remain are the lack of protective vaccines and the occurrence of asymptomatic individuals that sustain the transmission cycle. Moreover, knowledge on the exact immunological basis for the highly efficient trypanosome transmission and asymptomatic infection remains scarce. Following an infectious bite, inoculated metacyclic parasites rapidly adapt to the skin environment to establish a local infection and to continue a journey to systemic colonization. Making use of the tsetse fly vector, parasite reporter lines for fluorescent detection and in vivo bioluminescent imaging, immune-deficient mouse models and immunological profiling of parasite/saliva-exposed cells, we explored the role of innate immune responses in infection establishment and systemic colonization. Despite the armory of recruited anti-pathogen effector functions, parasites escape immune elimination and prominently distribute to tissues such as adipose, spleen and lungs. Similarly to most infectious diseases, the host immune system must maintain a delicate balance between mounting an effective immune response and limiting collateral damage, in which the role of IL-10 is paramount. Our studies have contributed to the characterization of two parasitic proteins with IL-10 inducing capacity promoting early trypanosome outgrowth in the host. Their pivotal role was illustrated by the reduced parasitemia and prolonged host survival upon gene disruption. Besides IL-10, interest has increased in the contribution of IL-27 as an alternative immune-modulating cytokine to control hyperactivation of IFN-g secreting CD4+ Th1 cells. Pharmacological inhibition and genetic IL-27-deficient models demonstrated an unexpectedly strong impact on both parasitemia and pathogenicity, revealing a prominent role of this cytokine during early establishment. Collectively, both IL-10 and IL-27 have been identified as pivotal cytokines triggered by trypanosomes, underlying successful establishment while limiting pathology. The discovery of asymptomatic colonization of the skin and lungs as tissue reservoirs pinpoints future challenges for disease control, but also offers opportunities for the development of novel non-invasive diagnostic tests.

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An analysis of phosphoproteome networks in resistant (BS90) and susceptible (BB02) snails of *Biomphalaria glabrata* shows immunoregulatory pathways.

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The freshwater snail *Biomphalaria glabrata* is the intermediate host of *Schistosoma mansoni*, a trematode parasite that causes the human disease schistosomiasis. In the current study, we analyzed the changes in protein phosphorylation, a well-studied post-translational modification, in resistant and susceptible *B. glabrata* strains to help us identify signaling pathways and protein functions modulated in the early stages after *S. mansoni* challenge. The phospho-protein profile of the head-foot region of sails exposed to schistosome miracidia for three hrs was analyzed using liquid chromatography with tandem mass spectrometry (LC-MS-MS) in resistant (BS90) and susceptible (BB02) strains, and compared to that of unexposed snails. Overall, we identified 1248 phosphorylated proteins, containing 2951 phosphorylated sites. Data analysis is undergoing using the gene ontology annotation software and the Kyoto Encyclopedia of Genes and Genomes (KEGG) to better understand the various categories and pathways associated with the response to parasite treatment. Preliminary results suggest that changes in phosphorylation levels in numerous proteins differ between resistant and susceptible snails. Interestingly, several of the proteins showing higher levels of phosphorylation after parasite treatment in resistant (BS90) belong to signal transduction pathways such as the wingless-related integration site (Wnt), the transforming growth factor-beta (TGF-β), and the mammalian target of rapamycin (Mtor) signaling pathways. Both the Wnt and TGF-beta signaling pathways have been found to be important for immune cell maintenance and function, suggesting that resistant snails are rapidly responding to the parasite presence and initiating a protective immune response; while the absence of these in the susceptible (BB02) snails may reflect a lack or delay of the immune response. Supporting a differential protein phosphorylation profile between strains, was the increased level of phosphorylation in a homologue of actin molecules associated with the phagocytic pathway in resistant compared to susceptible snails, corroborating previous studies that show increased phagocytosis activity in resistant snails compared to susceptible snails. This study will enable us to gain a greater understanding of the regulatory mechanisms involved in host-parasitic interactions, as well as identify some of the key signaling pathways involved in *Biomphalaria* snail's immune response.

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Helminth communities in migratory bird hosts of the family Turdidae.

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Due to federal protections for neotropical migrant songbirds, there has been a lack of recent research on their parasites and host specificity. However, building collisions provide a unique opportunity to study these topics. The objective of this research is to describe the community of ocular and intestinal helminth parasites that inhabit migratory bird hosts of the Turdidae family. Songbirds were collected during migration seasons of 2017-2018, and eyes and intestines were dissected to identify parasites. Six species of birds have been processed for eyeworms (n = 75) and intestinal helminths (n = 47). The veery, gray-cheeked thrush and wood thrush did not harbor any parasites. Five eyeworms (*Oxyspirura* sp.) were found in a single hermit thrush (n=33), and none were found in any other host species. The acanthocephalan *Plagiorhynchus* sp., or *Plagiorhynchus cylindraceus* when able to determine species, had a prevalence of 100% in American robins, 33% in hermit thrush, and 8% in Swainson's thrush. Cestodes had a prevalence of 75% in the robin, 8% in the hermit thrush, and 6% in the Swainson's thrush. Trematodes were identified as *Brachyamia* sp., thus far, with a prevalence of 4% in the hermit thrush, 18% in the Swainson's thrush, and none reported in the robin. The American robin was infected with an intestinal nematode: *Capillaria cf. contora*. While the hermit thrush was infected with several intestinal nematodes: *Porrocaecum ensicaudatum*, *Diplotriaena* sp., Spirurida, as well as a possible new species of *Capillaria*. The possible *Capillaria sp. nov.* is similar to *C. caudinflata* but has distinguishing characteristics such as shorter male caudal wings and coil-shaped bursal processes, as well as significantly shorter vulvar appendages in females. Overall, the acanthocephalan, *Plagiorhynchus*, was the most common helminth among all birds studied. The hermit thrush had the most diverse nematode parasites, including a new *Capillaria* species. The study sheds light on host specificity in Turdidae birds, but illustrates that more research in this field is needed.

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Feeding guild determines haemosporidian infection risk for waterfowl in the upper Midwest.

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Birds possess the most diverse assemblage of haemosporidian parasites including four genera, *Haemoproteus*, *Leucocytozoon*, *Parahaemoproteus*, and *Plasmodium*. Currently there are more than 250 morphologically identified avian haemosporidian species, although the true diversity is unknown, due to high genetic diversity and insufficient sampling across all avian host orders. Historically sampling has occurred predominately within passerines (songbirds), however recent work has highlighted the hidden diversity within non-passerine taxonomic groups, identifying distinct parasite clades within cranes and vultures. Additionally, non-passerine species demonstrate unique host ecological traits, which likely alter parasite diversity and transmission as has been demonstrated in passerines. Waterfowl (Order Anseriformes) are an ideal group to both discover hidden parasite diversity and to examine the role of host ecology in parasite transmission. Waterfowl contain two distinct feeding guilds, dabbling and diving ducks, which differ not only in feeding behavior but also in habitat utilization that likely alters vector encounter rates and haemosporidian parasite risk. To determine the role of feeding guild in haemosporidian parasitism we analyzed blood and liver samples collected from hunter donated waterfowl from North Dakota, Minnesota, and Wisconsin from 2019 to 2022. A total of 223 individual hosts, represented 21 host species, were screened for haemosporidian infection using a standard molecular approach targeting the barcoding region of the cytochrome b gene. A total of 93 individuals were infected by haemosporidian parasites (41.7% prevalence). Infection prevalence differed significantly between dabbling (56.9%, n=109) and diving (27.2%, n=114) ducks. In addition, each feeding guild supported a unique haemosporidian community, composed of different haemosporidian lineages. We found 10 novel lineages not before identified in any avian host, 4 *Leucocytozoon*, 2 *Parahaemoproteus*, and 3 *Plasmodium*. Phylogenetic reconstruction placed these lineages within clades known to infect waterfowl. These results demonstrate that waterfowl contain a diverse community of haemosporidian parasites, and that feeding guild alters parasite infection risk as well as parasite community dynamics. Studying non-passerine groups, such as waterfowl, is important to determine the true diversity of haemosporidian parasites and the role of host ecology in shaping parasite transmission.

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Haemosporidian diversity of Waterfowl in Green Bay, WI.

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Starting in the mid-1980s, Greater (*Aythya marila*) and Lesser (*Aythya affinis*) Scaup populations have been declining. Previous research focused on trematode and cestode diversity living in these hosts, linking major die-offs to non-native trematodes introduced in the Mississippi flyway. However, there is very little research on haemosporidian populations on waterfowl, particularly in diving ducks like Scaup. Our research objective is to survey haemosporidian prevalence and diversity in Scaup from Green Bay, WI. We obtained all birds for our study through hunter donations, totaling 67 Scaup and 15 other waterfowl representing seven additional species over the 2019-21 hunting seasons. These birds were dissected, and liver samples collected, preserved in 95% ethanol, and frozen. We extracted DNA from the liver samples and then amplified parasite DNA sequences using established protocols. This method allowed us to detect haemosporidian parasites and identify them to the genus level, including *Haemoproteus*, *Leucocytozoon*, *Parahaemoproteus*, and *Plasmodium*. Greater Scaup had lower overall haemosporidian prevalence (15.8%) than Lesser Scaup (27.6%). With both species combined, scaup had 18 total infections from 14 birds and the majority (61%) of parasites were *Leucocytozoon*. In all dual infections, at least one species of *Leucocytozoon* was detected. We discovered 4 new lineages: two from Scaup, and one each from a Wood duck (*Aix sponsa*) and a Common Goldeneye (*Bucephala clangula*). To further investigate patterns of diversity and host-associations we conducted phylogenetic analyses that revealed a lack of host specificity across waterfowl species. A generalized linear model showed that none of our demographic factors (species, age, sex, or year of collection) were statistically associated with parasite infection ($p>0.05$). Our findings may be representative of the larger scaup population that uses the Mississippi Flyway as previous studies indicate these parasites may be transferred at breeding grounds. Monitoring parasites in scaup is important for waterfowl management to better describe the distribution of potentially pathogenic species.

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Peculiar patterns of parasitism in the Lake Ontario Basin.

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This study highlights two parasitological puzzles that either directly or indirectly involve non-native Pacific salmon in Lake Ontario, New York. *Salmincola californiensis*, also known as gill maggot, is a lernaepodid copepod that is native to Pacific salmon in the Pacific Northwest in North America. It was introduced to the Ontario Basin sometime after its Pacific salmonid hosts were, and occurs there in *Oncorhynchus mykiss* (rainbow trout) at over double the prevalence that it does in its native range, and at a higher mean intensity. During the last three years we have observed a prevalence of 85% (2021), 75% (2022) and 88% (2023) at annual samples of 60 adult spawning fish in the Salmon River, an eastern tributary of the lake. In spite of high prevalence of *S. californiensis* in adult *O. mykiss* we did not encounter infections in younger age class fish, having examined 59 young of the year in 2019, and 30 juveniles between 2019 and 2021. These data suggest possible differences in transmission patterns of *S. californiensis* in the Ontario Basin compared to its native range. In the same eastern tributaries of Lake Ontario *Catostomus commersonii* (white sucker) were infected with two species of the acanthocephalan

genus *Neoechinorhynchus*, *Neoechinorhynchus bullocki*, which was previously reported from *C. commersonii*, and a second species of the genus that resembles Neoechinorhynchus salmonis, a species described from *Oncorhynchus nerka* (sockeye salmon) from British Columbia, Canada. The latter species of *Neoechinorhynchus* is evidently rare because we have only encountered it in four of 30 *C. commersonii* in the eastern tributaries of Lake Ontario, and in two of 134 *C. commersonii* in Oneida Lake, which is also part of the Ontario Basin. If those specimens actually are *N. salmonis* that made a host switch following their introduction, then we are puzzled by its lack of occurrence in the 306 salmonids of genus *Oncorhynchus* from our survey of the Ontario Basin. The other explanation, that the 2nd species of *Neoechinorhynchus* in *C. commersonii* is previously unreported and a new species, raises the obvious question of why it was never previously observed in this well-studied fish.

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Parasites are everywhere, but are they in museums? Move forward vouchering parasite specimens in the 21st century.

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For over two centuries, museums have been the center of scientific inquiry and education. They are one of the world's greatest treasures of information, and one of our most tangible links to the past. Numerous publications conclude that given rapid global change there is an urgency to voucher specimens and include parasites. Like parasites themselves, parasite collections must be adaptable over time to capture the diversity of parasitic taxa and associated host data. Parasitism, being a life-style rather than taxonomic distinction, includes a large swath of protozoan, metazoan, fungi, plant and other taxa. Practically this often means taxon specific preservation techniques, host and parasite vouchers and associated data. Although there is quick agreement among scientists about the value of museum collections and the need to fully voucher parasite diversity into museums, there has been relatively little discussion regarding the nuts and bolts of how this should be done in the case of parasites. One of the tenets of "good science" is that the study must be both repeatable and verifiable and a voucher is one of the most critical components in the scientific process. Vouchers allow for the verifiability of field studies involving the collection of specimens, behavioral studies using organisms with different taxonomic names. A review of the Journal of Parasitology over the last year shows that less than 40% of papers deposit voucher specimens and very few deposit host vouchers. Here we will focus primarily on heteroxenous metazoan parasites, though will discuss practices to voucher protozoan, prokaryotic and viral organisms, and present specific considerations and recommendations for vouchering parasitic taxa.

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Trematode species detection and quantification by environmental DNA-qPCR assay in Lake Chany, Russia.

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Environmental DNA (eDNA) surveys promise to be a sensitive and powerful tool for the detection of trematodes. This can contribute to the limited studies on trematode ecological contributions and connections, specifically in water ecosystems. Here, we applied the eDNA protocol to quantitatively detect trematodes and present the development of a novel eDNA qPCR assay of *Moliniella anceps*, *Opisthioglyphe ranae*, and *Plagiorchis multiglandularis* cercariae. Species-specific primers and probes were designed and assessed to quantify these trematode species. We evaluated the effectiveness of the assays using eDNA samples collected from different sites of Lake Fadikha and Kargat River Estuary in Lake Chany, Russia. Our data showed excellent specificity and sensitivity in all three assays. Further, all three assays have high efficiencies ranging from 94% to 105%. *Molinella anceps*, *O. ranae*, and *P. multiglandularis* were detected in the environmental water samples through real-time PCR. Thus, we anticipate that our developed assays would be beneficial for biomonitoring, measuring, and management of ecological systems.

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Parasites of the Northern Everglades, Florida fishes: Bangham, 1940 revisited.

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This is a project that will utilize parasitological data to inform us about environmental change. The study system is the northern Everglades and Lake Okeechobee in Florida. Lake Okeechobee has historically served as the headwaters of the Everglades within the Kissimmee-Okeechobee-Everglades ecosystem. This ecosystem has gone through significant changes that affect the water quality and hydrology since the late 1800’s, as well as species introductions and extirpations. A classic study by the parasitologist Bangham in the late 1930's provided parasite occurrence data from his survey of 1,380 individual fish representing 45 species throughout the region, including 56 parasite species that occurred at the adult stage in the digestive system of the fish. Bangham’s survey will serve as important baseline data for a system that has undergone extensive change since that time. Fish will be collected via electrofishing, seine, fish traps, and traditional angling. The complete digestive system of each fish will be examined for adult stages of parasites, but in some cases full necropsies will be conducted. Helminths encountered will be preserved using conventional methods for each major group, for both morphological and molecular study. Identifications to species of whole mounts of parasites prepared via conventional staining techniques will be done with the aid of light microscopy. Our initial goal is to determine how many of the 56 species of parasites that Bangham reported at the adult stage are still present in their respective fish hosts in this system. To attain this goal, we plan to examine 400 individual fish representing the 34 species of fish that were previously reported as hosts for adult stages of parasites. Given that there have been various environmental changes to the system, we predict that only a subset of the parasites reported by Bangham over 80 years ago are still present in the system today. The key result of this survey will be an updated host-parasite checklist; partial survey results will be provided in this poster presentation. The presence (or absence) of parasites will be interpreted in the context of the parasite life cycles in cases where those life cycles are known.

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A search for the missing fish parasites: A survey of the tributaries of Oneida Lake, New York.

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This study builds on a large ongoing survey of the parasites of Oneida Lake fishes. Oneida Lake is the largest in-state lake in New York. In 1932, Van Cleave and Mueller completed a 3-year study of the parasites of the fishes of Oneida Lake, in which they reported dozens of species of parasites. Since 1932, the lake has undergone many environmental changes which have impacted the fauna and flora of the lake. Although these changes are well-documented, there has only been a single published study on the fish parasites of Oneida Lake since 1932, and it focused on only two species of centrarchid fishes. The present study, along with the ongoing survey of the lake, aims to address how many of 65 species of parasitic worms originally reported by Van Cleave and Mueller have persisted in the Oneida system after its environmental changes. This poster presentation provides fish parasite survey data on a single tributary, Chittenango Creek, at an early juncture in this study, which will ultimately include data on multiple tributaries in the Oneida system. Fish were collected via backpack electroshocking. The digestive system of each fish was examined for parasites but in some cases whole necropsies were undertaken. Parasitic worms encountered were preserved for morphological study using conventional techniques and subsequently stained and mounted for light microscope study. The fish parasite survey results for Chittenango Creek are consistent with the ongoing survey results for the lake, with two notable exceptions. The trematode *Rhipidocotyle papillosa* was encountered in our small sample of only four *Micropterus dolomieu* whereas it was not encountered in the 16 *M. dolomieu* examined from the lake. Likewise, the trematode *Bunodera sacculata* was encountered in two of the 25 *Perca flavescens* examined in Chittenango Creek but not in the 88 *P. flavescens* examined from the lake. Both trematode species use a native clam as the first intermediate host. Native clams were displaced from the lake by invasive mussel species of *Dreissena* but our data suggest that these mollusks persist in the tributaries—a pattern that will be further investigated by extending our survey to additional tributaries.

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Linking horsehair worm presence with insect diversity: implications for ecosystem health.

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Nematomorpha, or the horsehair worm have complex parasitic life cycles. Their lives begin as eggs suspended in an aquatic freshwater environment. These eggs hatch into larvae that encyst in a paratenic host, which typically include the pre-metamorphosed larvae of aquatic insects, such as mosquitos, caddisflies, and mayflies. When these paratenic hosts become adults, they are consumed by the definitive host, which are typically terrestrial arthropods, such as crickets, roaches, or grasshoppers. Inside these hosts, the horsehair worms develop into juveniles. Once mature, they manipulate their host into entering water where they exit the host and mate. Previous studies have linked healthy, functioning ecosystems with abundant insect diversity. Such insect diversity provides horsehair worms with many hosts, paratenic or definitive, throughout its lifecycle. Therefore, we predict that horsehair worm presence at a site will correlate with high insect diversity. To test this, we collected adult caddisflies using a caddisfly attracting ultraviolet light trap at four sites. We collected other aquatic insects by hand and from four clay tiles placed at each site over 7 days. We hand collected twenty *Physa* snails and examined them for the presence of horsehair worm cysts. Finally, we measured nitrate, DO, pH, salinity,

and atrazine levels. One of the four sites tested positive for atrazine. Initial analysis showed two sites with cyst prevalences of 50% and 65% and cyst intensities of 21.6 and 2.7 respectively. We calculated insect biodiversity using the Shannon-Weiner diversity index and found indices of 1.13, 1.38, 1.33, and 0.37 at the sites. Correlation of these data will help us assess the relative health of each site. These data have the potential to forecast the consequences of climate change, agriculture, or other disruptions on aquatic ecosystems.

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Extrapolating large scale parasite data from public repositories.

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One pressing issue in modern parasitological studies is the difficulty in collecting large swathes of comprehensive data. While we might know where a parasite can be found or a large portion of its life-cycle, confidently assessing a complete picture of said life cycle or geographic range presents significant logistical issues which are very arduous to overcome. However, public repositories offer a unique method of supplementing data collection that can be implemented on a larger scale. These public repositories, such as Twitter or Google, can provide additional information such as new parasite sightings or population trends if such data is accessed and parsed properly. Utilizing the complete database of tweets from Twitter we compared its efficacy in modeling the trends found in the more dedicated parasitological database of iNaturalist by searching for occurrences of "*Nematomorpha*", "Hairworm" and "Gordian Worm" in the United States since 2007. These occurrences were compared seasonally (spring, summer, fall and winter) and geographically (east versus west coast of the U.S.). While Twitter data does not necessarily provide rigorous quantitative conclusions, tweets, and by extension public repositories, offer useful qualitative outcomes such as documenting sightings nationally and predicting promising locations to document new parasites.

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Parasites of coastal fish of Galveston Bay, Texas: Chandler, 1935 revisited.

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In 1935, Chandler conducted a fish parasite survey in Galveston Bay, Texas and encountered a diversity of parasites including ten species of adult helminths, nine of which were new species at the time. We were interested in finding which of these original species were still present at this location, with a focused interest in acquiring acanthocephalans for subsequent research. Our survey took place in July of 2022 and included fish collections via haul seine and traditional hook-and-line angling. In total we collected and examined 189 individuals representing 12 species of fish, seven of which were also part of Chandler's survey. Only 46 of the 189 individual fish examined were infected with adult helminths in the digestive system. Given that the diversity of trematodes, nematodes, and cestodes was underwhelming, we chose to focus on the acanthocephalans. In Chandler's original study three species of acanthocephalans were encountered, but only one, *A. verecundus* was found at the adult stage. Conversely, in our survey we encountered two species of acanthocephalans, both as adult worms. One of the species of acanthocephalans that we encountered was not observed by Chandler in his survey even though he looked at the same type of fish. *Floridosentis elongatus* was encountered in *Mugil cephalus* (striped mullet) in our survey. Fourteen of 65 *Cyprinodon variegatus* (sheepshead minnow) were infected with *Atactorhynchus verecundus*, a sample that included multiple individuals from the

Bolivar Peninsula, the type-locality of this Acanthocephalan. In addition, three of 18 *Fundulus grandis* (gulf killifish) were infected with *A. verecundus*, representing a new host record for this acanthocephalan. Conversely, no specimens of *Octospiniferoides chandleri* were present in the sample of *F. grandis* in Galveston Bay, Texas, the type-host and type-locality of that acanthocephalan.

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Intestinal helminths of the California ground squirrel (*Otospermophilus beecheyi*).

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California ground squirrels (*Otospermophilus beecheyi*) are important models for evolution, ecology, and animal behavior. The squirrels of the Briones Regional Park in the San Francisco Bay Area have been researched for over 15 years as a part of the Long-term Study of California Ground Squirrels. Squirrel colonies from distinct study sites are monitored for social behavior and life history traits, which are linked to the genetics and physiology of each tagged individual. Recently, ectoparasites (fleas) of these squirrels have been analyzed over multiple seasons and integrated into monitoring protocols. Wild rodents are typically rich with intestinal helminths, but the helminth community of California ground squirrels has not been surveyed for over 80 years. We have combined classical (fecal flotations) and cutting-edge (fecal eukaryote metabarcoding) parasitological approaches to define the intestinal helminths of these squirrels, setting a foundation for long term monitoring of many different parasitological features. Establishing the squirrel parasite community and developing species-specific barcoding primers will enable retrospective studies on banked fecal samples, empowering studies that link infection characteristics to a rich, long-term behavioral dataset. These data will also enable the development of new behavioral, epidemiological, and ecological hypotheses that can be tested using this unique model species.

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Genetic characterization of *Cryptosporidium* to study transmission dynamics in north India.

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Cryptosporidium, a protozoan parasite that infects epithelial cells of the gastrointestinal tract of vertebrates leads to the diarrheal disease cryptosporidiosis. There are two routes of transmission of this parasite: Anthroponotic and zoonotic. Studies on the transmission route of *Cryptosporidium* are limited from developing countries. In India, there is a lack of transmission data. We carried out a molecular analysis of *Cryptosporidium* from human, cattle, manure, vegetable and water samples to understand the zoonotic transmission in our region. A total of 500 fecal samples from persons suffering from diarrhea attending our referral institute in north India, and fresh 145 fecal samples from cattle and 10 manure samples were collected. Also 120, vegetable and 30 water samples were collected from surrounding regions. PCR was performed for three genes 18S rRNA, hsp70, gp60 followed by Sanger sequencing and further subtyped on the basis of TCA (A), TCG (G), TCT (T), and 5’ -AAA/G ACG GTG GTA AGG-3’ (R) repeats. Of the 500 human samples, 15 samples were found positive for 18SrRNA, hsp70 gene. In cattle and manure 5 samples were positive for *Cryptosporidium* spp., whereas two samples were positive in both water and vegetable samples. These samples were further subtyped into *Cryptosporidium* sub genotypes which belongs to various subfamilies of *Cryptosporidium hominis* and *parvum*. Subtypes of *Cryptosporidium* sp. found in the present study suggested anthroponotic as well as zoonotic transmission in this region. Zoonotic subtypes were also identified in water and vegetable samples which suggest of food borne and water borne transmission. However, more sampling needs to be done to assess the true status of zoonotic transmission of *Cryptosporidium.*

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Tritrichomonas foetus transcriptome: comparison of *in vivo* and *in vitro* profiles.

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Tritrichomonas foetus is an important pathogen of cattle with infections resulting in early embryonic loss and ultimately significant economic impact due to widespread calving intervals. This study aimed to analyze the transcriptome of experimentally infected heifer calves. A heifer calf model was experimentally infected with field strain trophozoites. Samples were collected weekly for 4 weeks for RNA extraction. Both *in vivo* and *in vitro* transcriptomes of the field strain trophozoites were compiled for comparison. A high level of similarity was found between the *in vitro* and *in vivo* transcriptomes. Our analysis provides important insights into diagnostic and vaccine candidate antigens. Additionally, this study supports the use of a heifer calf model for *T. foetus* infections in a research setting as infections were maintained for a significant period of time. Future studies should aim to compare the transcriptome of other *T. foetus* strains and longer-term infections.

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Prevalence of intestinal parasites and associated risk factors among inflammatory bowel disease suspected patients in Tigray Regional State, Northern Ethiopia.

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Background: Intestinal parasite infections are important public health concerns globally. Besides, some Intestinal parasite infections aggravate symptoms, have a clinical similarity, and considered differential diagnosis with Inflammatory Bowel Disease. So, the prevalence of intestinal parasites and associated risk factors among Inflammatory Bowel Disease suspected patients were determined. **Objective**: To determine the prevalence of intestinal parasites and associated risk factors among Inflammatory Bowel Disease suspected patients attending in Ayder Comprehensive Specialized Hospital, and Hiwot Private Clinic, Mekelle, Tigray, Ethiopia, 2020. **Methods**: A cross-sectional study was conducted among individuals who were Inflammatory Bowel Disease suspected patients in the Ayder Comprehensive Specialized Hospital, and Hiwot Private Clinic from February 01, 2019 to July 30, 2020. Descriptive statistics were computed to summarize data and result was presented using tables. Association between different variables with outcome was analyzed using Bivariate and multivariate logistic regressions. The p-value less than 0.05 were considered as statistically significant. **Results**: A total of 297 Inflammatory Bowel Disease suspected patients were included. Of these, 54.9% were males. The overall prevalence of intestinal parasites was 127 (42.76%). *Entamoeba histolytica/dispar* 76 (25.58%), and *Giardia lamblia* 32 (10.77%) were the most predominantly identified parasites. Participants with untrimmed fingernail (AOR =2.4 95% CI =1.3-4.3, p =0.002), eating unwashed vegetables (AOR=2.3, 95%, CI: 1.2-4.3, p =0.011), and family size of greater than five (AOR=1.7, 95% CI= 1.029-2.881, p = 0.039) were found to be independent

predictors of intestinal parasites. **Conclusion:** Overall prevalence of intestinal parasites infection was 127 (42.76%). *Entamoeba histolytica/dispar* were the most prevalent parasites. Family size of greater than five, eating unwashed vegetables, and untrimmed fingernail were found to be statistically associated with infection of intestinal parasites. Therefore, health care providers should screen and treat Inflammatory Bowel Disease suspected patients for intestinal parasites in order to ensure good diagnosis, and treatment.

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What's in a fish? A survey of the parasitic fauna of four freshwater sculpin (*Cottus* spp.) in Georgia.

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Cottus is a genus of nocturnal, predominantly freshwater fish that reside on the bottom of gravelly lakes and streams. There is a lack of knowledge pertaining to the parasitic taxa infecting the *Cottus* fish. Four species of *Cottus* were collected from freshwater streams in Georgia between 1999 and 2016, preserved in formalin and then ethanol, and stored in KSU's fish collection. We necropsied approximately 50 fish to collect their parasites; the gills and organs were removed from the fish to examine under a stereo microscope. We collected parasites for further investigation. In the *Cottus* specimens, we found nematodes, cestodes, acanthocephalans, and monogeneans. Our study is important because knowledge on types of parasites that are infecting freshwater sculpins is extremely limited.

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Ticks recovered from the nonnative Burmese python (*Python bivittatus*) in Southwest Florida.

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The introduction of nonnative species into Florida has become a pervasive problem as some species have now established their life cycles in the region. In addition to competing with native species for space and resources, these nonnative species introduce pathogens and parasites that could affect the health of native species. Already in Florida, nonnative species have introduced ticks that potentially carry pathogens. However, few studies have investigated the invasion of nonnative parasite species which is important to understand the effects on native species, but also the potential for zoonoses. In this study, we describe ticks recovered from the nonnative Burmese python collected from locations throughout South Florida. Ticks were received from The Conservancy of Southwest Florida during their annual collections. All ticks were found to be consistent with the morphology of *Amblyomma* sp. including a post-anal groove, mouthparts longer than basis capituli and scutum with eyes. DNA was extracted using Qiagen Blood and Tissue kits and then outsourced for sequencing. Species determination will be reported and their prevalence on Burmese pythons.

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Novel methodology for the study of *Toxocara canis* somatic larvae.

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Toxocara canis is a significant zoonotic pathogen, with human infections causing visceral larva migrans. The occurrence of somatic larvae and transplacental and lactogenic transmission pose serious challenges in eliminating this parasite from canines. Recovery of somatic larvae from hosts is tedious and has hindered the study of this life stage of *T. canis*. Our lab developed a novel method to study somatic larvae by utilizing an implantable device. This device contains the larval parasites while allowing for the exchange of host and parasite molecules. Our preliminary studies show *T. canis* larvae survival times of up to 10 weeks when implanted subcutaneously in mice. These findings support the further exploration of utilizing implants for the study of somatic *T. canis*. Future studies will aim to develop a model system for the study of hypobiotic larvae utilizing the implant. Additionally, this novel methodology could be employed to study life stages of a wide variety of parasites and life stages which are otherwise difficult to recover from experimental hosts.

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Aggression in male house crickets (*Acheta domesticus*) infected with hairworms.

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Parasite-induced behavioral alterations are common phenomena in hosts infected with parasites. While there are some behavioral alterations that facilitate transmission of the parasites to the next host, some behavioral alterations are simply by-products of infection. In some cases, these alterations are induced by changes in host physiology either directly or indirectly by the parasite. Within the phylum Nematomorpha, aquatic species of hairworms cause the host to enter water so the parasite can complete its life cycle. Beginning as microscopic larvae (~50 µm), hairworms increase their size by nearly 5000× within the cricket's abdomen. Although the water-seeking behavior of hosts infected with hairworms has been well-documented, few studies have examined other impacts on host behavior. In this study, we will determine the effects of parasitism on cricket host behavior including male aggression and locomotion. We will infect one hundred and fifty half-grown crickets by feeding crickets with snail tissue infected with the hairworm, *Paragordius varius*. Using established male cricket aggression protocols in a novel arena, a single, infected cricket will be paired with a size-matched uninfected cricket. This will be repeated with 30 sets of males. Each trial will run for three minutes and displays including aggressive chirps, mandible flaring, biting, and grappling times between opponents will be quantified. Aggressive behaviors measured are similar to Alexander (1961) and modified by Hofmann and Schildberger (2001). To determine mechanisms of behavioral alterations, the male gonads, post-trial, will be extracted, weighed, and measured. We predict infected males will display less aggressive behaviors compared to uninfected controls leading to uninfected crickets winning more often than infected crickets. We expect to find infected crickets have smaller gonads as a result of hairworms incurring damage to gonads compared to uninfected males. To record behaviors, we will use Noldus EthoVision XT, a video-tracking software. Recording behaviors will permit observers to more accurately record each behavior between crickets. This is the third study to examine behavioral differences between infected and uninfected male *A. domesticus*.

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Molecular characterization of human intestinal parasites in bivalves.

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Giardia lamblia, a flagellated protozoan parasite, if ingested, infects the lumen of the small intestines in humans. *G. lamblia* is most commonly transmitted to humans via ingestion of contaminated food and water. Its presence in marine environment is relatively unorthodox. The goal of this study is to determine the prevalence of *G. lamblia* in Atlantic oysters (*Crassostrea virginica*) and ribbed mussels (*Geukensia demissa*) collected from Orchard Beach and Clason Point, Bronx, NY. Each year, the mussels were collected in the fall. Tissues were dissected, followed by DNA extraction and PCR analysis. Thus far, we found a marked fluctuation in the prevalence of *G. lamblia* in both bivalves from 2014-2018, ranging from 4.5% to 42%, with no parasite detected in 2015 in the ribbed mussels collected from Orchard beach. In addition, a change in the genotype of *G. lamblia* was observed from year to year, with assemblage A being more frequent. Commercial bivalves were also examined for the presence of *G. lamblia*. A total of 30 oysters and 72 blue mussels (*Mytilis edulis*) specimens were purchased from a popular fish store located in the Bronx in the fall of 2019. *G. lamblia* DNA was detected in 20/30 of the oyster specimens with a resulting prevalence of 6.6%. On the other hand, *G. lamblia* DNA was detected in 9/72 blue mussels (12/5%). These data indicate that commercial bivalves are potential sources of *G. lamblia* infection. In conclusion, bivalves may be used as a bio-monitors of *G. lamblia*.

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Viability of snail-to-snail transfer of *Paragordius varius* cysts to crickets.

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Species in the parasitic phylum Nematomorpha, commonly called horsehair worms, are found in aquatic environments. Adults lay their eggs in water which hatch into larvae. Larvae encyst in the paratenic host, which are typically aquatic larval insects. After these hosts metamorphize into adults, they can be consumed by the definitive host where the nematomorph matures and eventually forces the host to enter water where the adult worm can emerge. *Paragordius varius* is a species of horsehair worm that is found in the waters of Nebraska where midges often serve as the paratenic host and crickets serve as the definitive host. Their larvae also encyst in aquatic snails. In this experiment, I explore whether aquatic snails can also serve as a secondary transfer host after *P. varius* had already encysted in a snail. To test this, I fed snails infected with *P. varius* cysts collected from a site near Lincoln, NE, to 31 uninfected tank-reared snails. Before they are fed to uninfected snails, the number of cysts in the infected snails was determined. After the uninfected snails have fed for several days in individual containers, they are examined, and the number of cysts in these snails was determined to calculate the transfer rate. Cyst counts were also conducted in the uninfected snails' frass 3 and 6 days after primary infection. Finally, the newly infected snails were fed to starved house crickets to determine if the cysts are still viable and will develop into adult *P. varius* after transfer through two host snails. If cysts can survive and mature while moving through two snail hosts, then follow-up studies should be done to determine the viability of cysts in other secondary transfer hosts and possible additions to the life cycle.

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Report of parasites from the gastrointestinal tract of cane toads (*Rhinella marina*) in Southwest Florida.

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Rhinella marina, known as the cane toad, is an invasive species introduced to Southwest Florida, originating from South and Central America. When invasive species, such as the cane toad, enter a new ecosystem, they bring along their parasites which are then released into their new habitat potentially infecting native species. In Southwest Florida, to our knowledge, no studies have examined the parasite fauna of the invasive cane toad nor examined the factors that affect the prevalence and size of parasites in their cane toad hosts. The objective of this study was to examine the prevalence and intensity of

parasites in the lungs and throughout the entire digestive tract of the invasive cane toads. Thirty cane toads were collected from SW Florida and returned to FGCU where necropsies were performed. The lungs and digestive tract were removed and inspected for parasites using a filtration method. When parasites were found they were removed, photographed, measured, and stored in 70% ethanol for later DNA analysis. The number of parasites were counted, and parasites were measured and noted if parasites came from male or female toads. We found several nematodes in the lungs of the toads in the genus *Rhabdias*. The most common parasites were nematodes followed by trematodes in the digestive tract. We found that host sizes correlated to the number of parasites present within the lungs and male vs. female correlated to the number of parasites present. Species identification will be discussed. This study provides the first description of parasites from the digestive system and lungs in cane toads in South Florida.

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Observations on turtle acanthocephalan abnormalities from Oklahoma, U.S.A.

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Adult acanthocephalans in the genus *Neoechinorhynchus* are commonly found infecting a wide range of turtle definitive hosts throughout North America. Other than reports on basic information such as intensity and species, little information exists on abnormal occurrences of acanthocephalans in turtles. We present four unusual cases of turtle acanthocephalans interacting with the host and other turtle parasites. Red-eared slider turtles (*Trachemys scripta elegans*) were surveyed for helminth parasites in Oklahoma, U.S.A. during 2010–2022. We observed 1) an acanthocephalan tied in a knot inside the turtle intestine, 2) an acanthocephalan attached with its proboscis to the outer serosa layer of the turtle intestine, 3) acanthocephalans embedded in the tissue of adult trematodes in the turtle intestine, and 4) nematodes attached to the tissue of acanthocephalans in the turtle intestine. These reports highlight some rare, but interesting, observations on adult acanthocephalans in their turtle hosts.

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Prevalence and diversity of gastrointestinal parasites associated with diarrhea in buffalo calves under extensive Buffalo farming in Bangladesh.

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Buffalo calves are the future for livestock industry. Calf care is not only essential for sustenance of the dairy industry but essential for quality germ plasm in preserving and maintaining good trait. Buffalo calves are one of the most popular ruminants reared in extensive condition for basically milk and meat. Gastrointestinal (GI) parasitism in calves possesses a critical warning factor that severely disrupts the growth performance. Effects of GI parasites may vary with age, sex of calves, nutritional condition and severity of infection and rearing practices. Thus, the current study was aimed to assess the prevalence and diversity of GI parasites in buffalo calves in Bangladesh. The fresh fecal samples (n=75) of buffalo calves suffering from diarrhea were collected and immediately preserved in cool conditions. These samples were processed via direct wet mount, sedimentation and floatation methods and examined under a light microscope. The results showed an overall 100% prevalence of GI parasites in the buffalo calves with diarrhea. Among these, *Toxocara*, *Cryptosporidium*, *Eimeria*, *Balantidium coli*, *Fasciola* and

amphistomes were the most prevalent parasites in calves suffering from diarrhea. One calf died due to severe GI parasitic infection that revealed after post-mortem. The higher prevalence and greater diversity of GI parasites indicate the association with the calf rearing husbandry practices under extensive buffalo farming condition in Bangladesh. Sharing of the same pastureland and water bodies by buffaloes significantly increases the prevalence of GI infection via interspecific transmission. The existing practice of rearing calf and adult buffaloes and other livestock within the same shed pose the potential risk of diarrheal pathogen transmission. Therefore, training to the local buffalo farmers for healthy husbandry practices and therapeutic strategies for buffalo calves should be provided to reduce the cross-transmission of potential diarrheal parasites and calf mortality for profitable buffalo farming in Bangladesh.

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Chordodes morgani effect on host growth and its development time in differing diet and temperature conditions.

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Chordodes morgani [Nematomorpha] is found throughout Nebraska and typically uses mayflies as its aquatic paratenic hosts and wood roaches (*Parcoblatta* spp.) as its definitive terrestrial host. Infection with nematomorphs entails energy drains on their hosts, which may slow the growth rate of the wood roaches. Further, lab infections with *C. morgani* took approximately 3 months to yield adult worms. Yet, in the field, adult *C. morgani* worms are observed only once a year (July), suggesting a 9 to 10-month development time in conditions with limited food and low temperatures. To simultaneously examine *C. morgani* developmental time and host growth rates, we designed a block experiment. Wood roaches infected with *C. morgani* were split between a cold treatment to simulate winter and a room temperature treatment to simulate lab conditions. Each group was further split into an unlimited food treatment and a restricted food treatment. A control group of uninfected roaches were split into the same treatments. All wood roaches were individually housed, and their lengths and weights were measured each week. Only 31 of the 106 roaches survived after 37 weeks. At this time, the remaining roaches were dissected, and no worms were found among either the uninfected or infected treatments. Results are discussed in the context of future directions.

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Risk Factors for *Strongylus vulgaris* Infections in Grazing Horses in Sweden.

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The equine parasite *Strongylus vulgaris* is a significant cause of disease in grazing horses and ponies worldwide and is particularly pathogenic due to its ability to cause fatal vascular damage. The parasite's life cycle takes approximately six to seven months to complete, and horses become infected by ingesting the third larval stage while grazing. Although an intense deworming program have reduced the prevalence of *S. vulgaris*, it has also led to widespread resistance to anthelmintics in small strongyles. Currently, 40-60% of horse establishments have at least one horse infected with *S. vulgaris* in Sweden, and there is a risk of neglect and untreated infections if extended egg hatching analyses are not performed. This study aimed to identify potential risk factors for *S. vulgaris* infection and develop detailed recommendations for further parasite control. An internet questionnaire was distributed to Swedish stable owners, of which 1278 stables finished the full questionnaire, and they were distributed throughout Sweden. Sweden's selective anthelmintic treatment rules require diagnosis before treatment and offer a unique chance to study factors affecting prevalence. The results showed a significant association between the occurrence of *S. vulgaris* on stables with more than eleven horses, as well as stables with grazing areas smaller than 0.5 hectares (1.25 acres) per horse. In addition, stables with concurrent sampling and deworming routines, as well as those that dewormed new horses before arrival to the stable, had a lower occurrence of *S. vulgaris*. However, the occurrence of *S. vulgaris* was not significantly affected by other factors such as pasture hygiene practices, permanent pastures, horses per pasture, type of pasture, housing system, or quarantine for new horses. We have found several factors associated with *S. vulgaris*. Informing animal owners about best deworming practices and preventive measures are important tools for reducing the buildup of anthelmintic resistance for small strongyles as well as reduce anthelmintics use.

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Case report: Severe infestation with tungiasis in a coastal community in Badagry Lagos, Nigeria.

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Tungiasis is an ectoparasitic infection caused by the invasion of the skin of a man, dog, pig, or bird with gravid female *Tunga* species. Prevalence may reach up to 50% in impoverished areas of South America, the Caribbean, and Sub-Saharan Africa. Very severe cases characterized by more than 30 embedded sand fleas may be found but are not common. Few studies have been done in Nigeria, and in endemic communities of Lagos, a prevalence as high as 45.2% has been reported. We hereby report a severe case of Tungiasis in Ilaje, an endemic community in the Badagry Local Government area of Lagos during an active surveillance conducted between August 2021 to May 2022 involving Eighteen (18) communities. This case is a 54-year-old male farmer who presented with pain, inflammation, ulceration, and deformation of digits, heels, and soles of his limbs. He reported a burning sensation and pruritus and was observed to have difficulties in walking, evidenced by a noticeable totter. Embedded fleas in clusters numbering between 30 to 60 per limb were extracted by manual removal and the wounds were treated topically. Tungiasis morbidity often leads to poor health and impoverishment. Although tungiasis is rare in nonendemic areas, however, with the increased travel and opening up of Badagry as a seaport and tourist site, this ectoparasite may further be disseminated if still neglected. It is therefore imperative to investigate the occurrence, create awareness, and implementation of preventive measures among local inhabitants. Physicians should be familiarized with this infestation for easy diagnosis and treatment.

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Alternative definitive hosts for the horsehair worm *Chordodes morgani*.

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Horsehair worms [Phylum Nematomorpha] lay their eggs in water where they hatch into larvae that encyst in aquatic insects. After the insect metamorphizes into an adult, it transfers the cyst stage to the terrestrial environment where an arthropod definitive host consumes it. The parasite then matures, driving this host to find water so the parasite can emerge in water as a free-living adult. Previous studies have shown that the nematomorph, *Chordodes morgani*, uses mayfly larvae as its aquatic host and wood roaches (*Parcoblatta spp*) as its definitive host. The goal of this experiment is to determine if other closely related insects could act as definitive hosts for *C. morgani*. To test this, we exposed seven mantids (*Stagmomantis carolina*) to live mayflies presumed to harbor *C. morgani* cysts in July 2022. We also exposed four American roaches (*Periplaneta americana*), and 29 Dubia roaches (*Blaptica dubia*) to *C. morgani* cysts. Insects were reared until they died whereupon they were dissected and examined for worms. After dissection, no roaches or mantids were found to contain any horsehair worms. Since these closely related insects cannot serve as a definitive host for *C. morgani*, then this horsehair worm species exhibits high definitive host specificity and may be more vulnerable to environmental changes than previously thought. More research needs to be done to confirm this and determine if this is true for other species of Nematomorpha.

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Echinococcus multilocularis: Is the European haplotype endemic in the Eastern US?

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The tapeworm *Echinococcus multilocularis* is a zoonotic parasite of public health importance. Wild canids and domestic dogs represent the main definitive hosts, in which the adult worms reside as an asymptomatic intestinal infection. In humans, the larval stages of *E. multilocularis* cause alveolar echinococcosis (AE), characterized by severe and progressive disease of the liver similar to malignancy, with possible metastases to other organs. Without treatment, the disease is lethal. In the US, the North American strain of *E. multilocularis* has traditionally been considered endemic to wildlife in Alaska and several northern midwestern states. Recent human and canid cases with the more pathogenic European haplotype of *E. multilocularis* in the Eastern US suggest the parasite is emerging and endemic in this new geographic location. Since 2019, we have tested 1,102 wild canids from Virginia, Pennsylvania, and Vermont. Fecal samples were collected from fox and coyote carcasses provided by the Virginia Department of Wildlife Resources, the Wildlife Future Program in Pennsylvania, and the Vermont Fish & Wildlife Department. No IACUC protocol was needed for this project. Samples were tested by a commercial qPCR test for *Echinococcus* spp. and *E. multilocularis* (IDEXX Laboratories) and a PCR test for *E. multilocularis* (Virginia Tech). Here, we are presenting three new cases of *E. multilocularis* in wild canids from Virginia and Pennsylvania. Although mitochondrial sequencing is underway to determine the origin of these new isolates, completed *nad2* sequencing is consistent with the European haplotype. Veterinarians should be aware of the presence of this tapeworm in the area and consider it on their list of differential diagnoses. Mapping the extent and prevalence of the European haplotype of *E. multilocularis* in US wildlife will help to better understand the risk to humans.

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Birds can serve as a dispersal agent for *Gyrinicola batrachiensis* (Oxyuroidea: Phayngodonidae) thick-shelled eggs.

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The oxyurid nematode *Gyrinicola batrachiensis* infects the gastrointestinal tract of tadpoles, and female worms are known to have a complex reproductive strategy wherein thin-shelled eggs are autoinfective and thick-shelled eggs are defecated into the aquatic environment to infect other tadpoles. The worms cannot infect adult frogs. While several recent studies have addressed different aspects of this reproductive system, no studies have explored survivorship of the thick-shelled eggs in different environmental conditions. The eggs are known to be viable for at least a few days in pond water, however it is unclear how this pinworm reestablishes infections after an isolated wetland dries and refills. Two mechanisms were tested to determine how pinworms re-establish infections in ephemeral wetlands: (1) Whether thick-shelled eggs could survive transport through a bird's (*Gallus gallus* or *Anas platyrhynchos*) gastrointestinal tract, or (2) if the eggs can survive buried in a moist soil environment that simulated a dried wetland. Birds were fed infected tadpoles, then feces were collected and searched for pinworm eggs. A bird's stomach environment was also simulated using pepsin to determine the effects of low pH on egg survival. Finally, eggs were layered in moist paper towel and buried to determine if they could survive for various time intervals. All surviving eggs were tested for viability at the end of each experiment by exposure to uninfected tadpoles and subsequent dissection. Overall, our results suggest that thick-shelled eggs have poor survivability in moist soil conditions, but are able to survive low pH environments, including passage through the gastrointestinal tract of a bird. We suggest that these pinworms can be introduced to temporary, isolated wetlands by birds that feed on infected tadpoles and defecate at multiple wetlands.

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Artemisinin based combination therapy failure in complicated *Plasmodium falciparum* malaria with concurrent enteric fever: a case report from North India.

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Artemisinin derivatives are the most efficacious parasiticidal choices of all anti-malarial drugs against complicated *Plasmodium falciparum* malaria. As per WHO/NVBDCP guidelines for complicated (cerebral) malaria, ACT is used as the first line treatment modality. Artemisinin based treatment failure in malaria patients in endemic regions can be attributed to parasite resistance to this treatment. Over the last two decades, artemisinin resistant strains of *P. falciparum* have emerged and spread in India and surrounding South-East Asian countries which may lead to increased malaria burden in these regions. Here, we report a case from North India with ACT treatment failure. A middle-aged man from New Delhi presented to our tertiary care hospital with acute febrile illness with altered mentation, loss of consciousness and convulsions and was diagnosed with *P. falciparum* malaria with cerebral complications and received ACT as an inpatient. Despite receiving appropriate treatment as per guidelines, the patient deteriorated and showed signs of late clinical failure. Since common causes of acute febrile illness present with non-specific and overlapping signs and symptoms, further investigations were also done for ruling out other etiologies of AFI. Rising titers were seen on Widal test

and liver function tests were severely deranged which may have contributed to the subject's poor prognosis. Molecular sequencing was performed to look for genes associated with drug resistance - which revealed a single mutation in *pfdhps* gene and double mutations in *pfcrt*, *pfdhfr* and *pfmdr-1* genes - that could have resulted in treatment failure. The subject eventually expired due to complicated cerebral malaria with multi-organ dysfunction syndrome and dyselectrolytemia. Our case report sheds light on the need for rigorous monitoring of signs of treatment failure among *P. falciparum* patients and the role of gene mutations in parasite response to anti-malarial drugs. It also highlights the need to correctly identify the cause of acute febrile illness – which may be challenging due to subclinical infections, previous infections, or co-infections.

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A method for isolating and sequencing trypanosome cells to investigate species associations in multiple morphotype infections.

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Trypanosomes are a diverse group of flagellated protozoa that are typically vector-borne and can infect the circulatory system of all classes of vertebrates. Amphibians are common hosts for trypanosomes and are often infected by multiple trypanosome morphologies in the bloodstream. These mixed infections complicate studying amphibian trypanosome diversity because sequencing mixed infections yield messy chromatograms, indicating that more than one genotype is present. Therefore, specific methods are needed to link sequence data to bloodstream trypanosome morphology. In this study, we detail the isolation and sequencing of individual trypanosome cells from frog blood for phylogenetic investigations of amphibian trypanosome diversity. Single trypanosome cells were isolated by dilution from frog blood containing multiple trypanosome morphotypes and three DNA extraction methods and five nested PCR primer regimes were utilized to optimize amplification from very low starting DNA concentrations. Isolated cell morphology was observed with a microscope prior to PCR, linking morphology to sequence data. DNA extraction methods included spin columns, no extraction initial hot-cold PCR cycles, and a Direct PCR kit. The nested PCR regimes targeted the 18s rRNA and glycosomal GAPDH genes, both separately and simultaneously with multiplex PCR. Although the success rate varied in the different combinations of extraction methods and primer regimes (29% to 50% with the use of a Direct PCR kit having the highest success rate), multiple individuals of all 6 trypanosome morphotypes were sequenced for both genes in a novel way that linked sequence data to cell morphology. Additionally, all 6 morphologically distinguishable morphotypes coinfecting a frog were genetically distinct, offering interesting insight into the assumption that trypanosomes often develop through different bloodstream stages. The methods presented here offer a promising solution to overcome the difficulties posed by multiple morphotype infections in amphibian trypanosomes.

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Improving diagnostics of *Crassiphiala bulboglossa*, the type-species of *Crassiphiala* and a common causative agent for black spot disease in fishes.

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Crassiphiala is a genus of diplostomid digeneans parasitic in kingfishers throughout the Western hemisphere. Its members use fishes as second intermediate hosts, and are commonly associated with the black spot disease in a variety of fishes. The type-species, *Crassiphiala bulboglossa*, was originally described by Van Haitsma in 1925 based on material from belted kingfishers collected in Michigan. Many descriptions of adult diplostomids have been based on laterally oriented and contracted specimens. The description by Van Haitsma is no exception and lacked measurements of many features commonly used in diplostomid taxonomy. No prior study sequenced morphologically identified *C. bulboglossa*. In the present study, we collected new specimens of *C. bulboglossa* from a belted kingfisher in North Dakota. Initial examination of specimens revealed several morphotypes with seemingly substantial differences. A fragment of the cytochrome *c* oxidase I (cox1) mtDNA gene was sequenced from hologenophores and vouchers of different morphotypes. No genetic differences were detected among individuals. Based on the combination of molecular and morphological data, we provide descriptions and illustrations of each morphotype based on both ventro-dorsally and laterally oriented specimens. The combination of our new DNA sequences and detailed morphological data of *C. bulboglossa* provide substantially enhanced diagnostic tools for this pathogenic digenean. This study was funded in part by the National Science Foundation (grant 1852459), the National Institutes of Health (IDeA grant number P20GM103442), and the University System of Georgia STEM Initiative IV.

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Systematics and phylogeography of fleas in the subfamily Doratopsyllinae (Siphonaptera: Hystrichopsyllidae).

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Doratopsylline fleas are widely distributed as parasites of the shrew family Soricidae in the genera *Sorex*, *Blarina*, and *Cryptotis* throughout northern hemisphere temperate zones. Two flea genera in this subfamily occur in North America, the Holarctic genus *Doratopsylla*, a monotypic genus found on shrews in the eastern US, Canada, and Eurasia, and the primarily Nearctic genus *Corrodopsylla*, including 3 described species in the western US, Central America, and Canada, as well as one species in Eurasia. The focus of this research is to generate a multi-locus phylogeny of representatives of all recognized species of the genus *Corrodopsylla*: the single Eurasian species, *Corrodopsylla birulai*, and all New World species of the genus *Corrodopsylla* currently recognized by Morrone and Acosta 2006, including *Corrodopsylla barreri* Traub & Evans, 1967 and *Corrodopsylla lira* (Traub, 1950) from Central America. Outgroup taxa include *Doratopsylla blarinae* and *Ctenophthalmus pseudagyrtes*, the latter identified as a sister clade of the Doratopsyllinae by Whiting et al. 2008. In addition to DNA extraction, flea exoskeletons are preserved and slide-mounted (Durden et al. 2020), identified morphologically, digitally imaged, and deposited in the Museum of Southwestern Biology as permanent voucher specimens to enable extended research on the phylogeography and systematics of this group to compare with similar studies of host phylogeography. This research will contribute to ongoing efforts to understand the complexity of faunal diversification events related to climatic cycling in the northern hemisphere which have impacted mammalian host/parasite dynamics across space and time. Project funding provided by supplement to NSF (DBI 19001920), Terrestrial Parasite Tracker (TPT).

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Imaging parasites using variable pressure scanning electron microscopy.

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Scanning electron microscopy (SEM) is used by parasitologists to study the anatomical landmarks of various parasites. Conventional SEM requires drying of the specimen and coating them with a conductive material before viewing the specimen in a near vacuum. Specialized equipment is needed to apply the conductive material, and without the coating, specimens would exhibit charging that impairs visualization of the specimens. An alternative method of imaging is variable pressure SEM (VP-SEM), which leaves some air in the image chamber. Consequently, the individual air molecules remove the charging from specimen and images can be obtained. Thus, specimens do not require coating with a conductive material prior to imaging. This study will assess the feasibility of obtaining useable images of parasites with VP-SEM. Specimens collected during previous research projects were used in this project. In preparation for VP-SEM, specimens were dehydrated through an ethanol series with a final dehydration step in hexamethyldisilazane (HMDS). Specimens were air dried and mounted to double-sided carbon tape on aluminum stubs. Specimens were then imaged under low-vacuum VP-SEM with a goal of imaging structures not typically observed in light microscopy. Images from VP-SEM were then compared to published, conventional SEM images of similar species to determine if VP-SEM could be a suitable alternative to conventional SEM.

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A comparison of parasites in the digestive tracts of the invasive Burmese python (*Python bivittatus*) compared to native snakes in Southwest Florida.

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The state of Florida is notorious for the introduction of exotic reptiles and amphibians. Many of these animals were brought to Florida through the exotic pet trade, most notably, the Burmese Python. Pythons have established in South Florida and directly affect the ecosystem by predating on native wildlife, but can also cause indirect issues such as introduction of novel diseases and parasites. Parasites such as *Raillietiella orientalis* were introduced into Florida's ecosystem by the Burmese pythons. It is now documented that *R. orientalis* has begun infecting native snakes, extending the range of the parasite beyond the range of pythons. However, no studies have examined other parasite fauna within Burmese pythons. In this study, we sampled native road-killed snakes and invasive snakes from Collier County, Florida. We performed necropsies to sample the parasitic communities within them. We found high prevalence and intensities of parasites that included cestodes, nematodes, trematodes, and pentastomes in native snakes. In pythons, however, only nematodes were observed indicating that their parasite fauna are not as species rich. This suggests that exotic hosts are not as susceptible to native parasites, indicating spillback (exotic species receiving local parasites) occurs less frequently than spillover (exotic releasing parasites into native species). This complicates management and conservation strategies for native Florida wildlife because Burmese pythons are thriving in South Florida. To our knowledge, this is the first study to examine the parasite fauna of the invasive Burmese pythons in South Florida.

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Rhinebothriideans of two species in the stingray genus *Hypanus.*

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Four species of stingrays in the genus *Hypanus* are known to inhabit the northwestern Atlantic Ocean, including the Gulf of Mexico. Although the tapeworm faunas of the southern stingray (*Hypanus americanus*) and the longnose stingray (*Hypanus guttatus*) have received some attention, tapeworms parasitizing the Atlantic stingray (*Hypanus sabinus*) and the bluntnose stingray (*Hypanus say*) are far less well known. Published reports of the tapeworms of *H. americanus* and *H. guttatus* indicate that members of the order Rhinebothriidea are particularly well represented by species of *Rhinebothrium* and *Anthocephalum*. The primary goal of this study was to increase our knowledge of the rhinebothriidean tapeworms parasitizing species of *Hypanus* to evaluate faunal patterns across members of the genus more broadly. The secondary goal was to morphologically characterize rhinebothriidean species included in published works on tapeworm mitogenomes and work in progress using targeted gene capture. Tapeworms studied came from five individuals of *H. sabinus* and nine individuals of *H. say* collected between 2005 and 2015 in the northern Gulf of Mexico and off the coast of South Carolina. Examination of specimens using light and scanning electron microscopy yielded an unexpected amount of diversity in the rhinebothriidean families Escherbothriidae and Rhinebothriidae. Not surprisingly, congeners parasitizing the same host species were the rule rather than the exception. *Hypanus sabinus* was found to host two species of *Scalithrium*, one species each of *Stillabothrium* and *Rhinebothrium*, and two species provisionally placed in *Spongiobothrium*. *Hypanus say* was found to host three species of *Scalithrium*, one species of *Rhabdodobothrium*, three species of *Rhinebothrium*, and two species provisionally placed in *Spongiobothrium*. At least one species of *Scalithrium* appears to be shared by both host species. This work highlights the importance of focusing more attention on the distinctions among closely related rhinebothriidean genera.

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Of intestinal helminths and *Mephitis mephitis*: The importance of DNA barcoding.

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Seventy-eight striped skunks (*Mephitis mephitis*) from southern Illinois were examined for gastrointestinal parasites. The helminthological record consists of 9 species, including the digenean *Alaria taxidea*, the cestodes *Mesocestoides corti* and *Oshmarenia mephitis*, the acanthocephalans *Macracanthorhynchus ingens* and *Oncicola canis*, and the nematodes *Baylisascaris columnaris, Molineus barbatus, Physaloptera maxillaris* and *Placoconus lotoris*. An unidentified species of acanthocephalan and *Dipetalonema* spp. were recovered from the body cavity. The most abundant and prevalent gastrointestinal parasite species found was *P. maxillaris* at 90%, followed by *P. lotoris* and *B. columnaris* both at 30%. We documented 3 additional species to the previous helminthological record, which includes *O. canis, M. barbatus* and *Dipetalonema* sp. Furthermore, 5 of the species found are shared with other carnivores present in the region. We propose to generate barcodes for each of these species to enable the monitoring and surveillance of these pathogens. This analysis is important because it helps us to see if there are any occurrences of coinfections with microparasites, including babesiosis and canine distemper virus.

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Parasites of fishes in streams of Soberania National Park, Chagres River drainage, Panama.

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Earlier parasitological surveys, between 2010 and 2019, of 15 species of fishes belonging to 8 families from tributaries of the Chagres River system in Panama, have shown that, with some notable exceptions, their parasite fauna is arguably an extension of neotropical source faunas farther south. In January of 2023, we returned to these streams and examined 111 fish from Quebrada Juan Grande and Rio Frijolito, representing 12 families and 18 species as follows: Characidae (4 species); Curimatidae (1 species), Erythrinidae (1 species), Lebiasinidae (1 species), Heptapteridae (2 species), Loricariidae (1 species), Trichomycteridae (1 species), Hypopomidae (1 species), Eleotridae (1 species), Gobiidae: (1 species), Cichlidae (3 species), and Poeciliidae (1 species). In total, 31 species of parasites were collected, namely Opalinidae (1 species), Monogenea (2 species), Digenea (13 species, 8 as adults), Nematoda (13 species; 11 as adults), Crustacea (1 species) and Myxozoa (1 species). Notably, no cestodes or acanthocephalans were found in the 2023 survey. In previous surveys we found *Quadrigyrus* sp. in *Hoplias microlepis,* the introduced Asian tapeworm, *Schyzocotyle acheilognathi* in cichlids, and immature individuals of a proteocephalid tapeworm in *Rhamdia quelen*. A preliminary assessment of the collection indicates several new records, including potentially new species (4 trematodes and 2 nematodes). Fish species in this (2023) survey that were also previously examined between 2010 and 2019 had the same parasite assemblages as before. *Cyphocharax magdalenae* (Curimatidae), *Awaous banana* (Gobiidae) and *Trichomycterus striatus* (Trichomycteridae) were examined for the first time, yielding two new nematode records; the presence of the nematode *Cosmoxynema* sp. (Pharyngodonidae) in *Cyphocharax magdalenae* extends the range of this nematode genus into Central America. Overall, the endohelminth fauna continues to display host specificity at the fish family level, clear neotropical affinities, and is dominated by trematodes and nematodes.

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No longer chambers of secrets: Characterizing the anatomy and mucosal surface of the spiral intestines of batoid hosts belonging to "tetraphyllidean" cestode Clade 4.

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In theory, the complexity of elasmobranch spiral intestines provides a broad spectrum of habitats for varieties of cestode groups. But, in reality, little is known about the anatomy or mucosal configuration of the majority of elasmobranch groups. The goal of this study is to characterize the spiral intestines of the sting ray and guitarfish hosts of cestodes belonging to Clade 4—a cestode clade currently included among the non-monophyletic order "Tetraphyllidea." Elasmobranchs examined consisted of 9 species in the stingray family Dasyatidae, 2 species in the bat ray family Myliobatidae, 2 species the guitarfish family Glaucostegidae, 2 species in the sawfish family Pristidae, and 3 species in the wedgefish family Rhinidae. Gross examination of spiral intestines was conducted to evaluate overall anatomy and to determine gross variation in the mucosal surface of the numerous intestinal chambers across the length of the spiral intestine. Mucosal surfaces were initially characterized with fast green. Samples of intestinal chambers exhibiting different mucosal features were subsequently collected and either examined in more detail using histological sections or prepared for and examined using scanning

electron microscopy. In all species, examined elements of the mucosal surface were most pronounced in the anterior chambers of the spiral intestine and became less pronounced along the length of the spiral intestine. In general, the mucosal surfaces of myliobatids exhibit regular anastomosing ridges. Those of dasyatids exhibit anastomosing ridges or pits bearing irregularly shaped or elongate villi. The mucosal surfaces of rhinopristiforms consist of anastomosing ridges with wide, irregularly shaped villi. Rhinopristiforms also have abbreviated flaps in between larger flaps, decreasing in number posteriorly. In Clade 4, three basic scolex morphologies are observed: those with bothridia that are foliose, hollow, or pouch-like. Taxa with the foliose bothridia are found in myliobatids. Those with hollow bothridia are found in rhinids. Those with pouch-like bothridia parasitize dasyatids, glaucostegids, myliobatids, rhinids, and pristids. Further work with *in situ* cestodes specimens will provide insights to test the relationship between scolex morphology, size, and attachment to host gut mucosa.

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Trichomonas tenax stimulates HL-60 neutrophil chemotaxis *in vitro.*

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Trichomonas tenax is a protozoan implicated in periodontal disease. Neutrophils play roles in parasite clearance and inflammatory pathology, but little is known about the chemical cues that affect neutrophil migration during oral trichomoniasis. Here, we determined whether *T. tenax* cell lysates or excretory-secretory products (ESPs) induce chemotaxis in human HL-60 neutrophils. To produce ESPs, *T. tenax* cells harvested from TYM medium were washed twice in serum-free RPMI (SFRPMI) and suspended in SFRPMI at 10^6 live trophozoites/ml. The cells were cultured for 24 h at 37°C in 5% CO_2, pelleted by centrifugation, and the ESP-containing supernatants retained for assays. For lysates, *T. tenax* cells in SFRPMI were disrupted by sonication, centrifuged to remove debris, the supernatant passed through a 0.2 µm filter, and the volume adjusted to the equivalent of 10^6 *T. tenax*/ml. HL-60 cells were differentiated to neutrophils in complete RPMI medium containing 1.2% DMSO for 5 days at 37°C in 5% CO_2. Differentiated HL-60 (dHL-60) cells were suspended in SFRPMI at 10^6 cells/ml. Transwell inserts (6.5 mm diameter, 3 µm pore size) were loaded with 0.2 ml/insert of dHL-60 suspension and positioned over bottom chambers containing 0.6 ml/well ESPs, lysates, 10% FBS in RPMI (positive control), or SFRPMI (negative control). After incubation for 3 and 6 h, migrated cells in bottom chambers were stained and counted. Experiments were repeated on four separate days with triplicate replications for each condition. Results were compared by one-way ANOVA and the Holm-Sidak post test, and were reported as mean migrated cells ± SD. Incubation time had no effect on dHL-60 migration; migrated cell counts for a given treatment were essentially the same after 3 or 6 h of incubation ($p>0.05$). Migrated cell counts induced by ESPs, lysates, or FBS were all significantly higher than that induced by SFRPMI ($p<0.001$). Migration induced by FBS ($4.13\pm0.37 \times 10^4$ migrated cells) was significantly higher than that induced by ESPs ($1.64\pm0.42 \times 10^4$ cells) or lysates ($1.26\pm0.21 \times 10^4$ cells) ($p<0.001$). ESPs induced significantly more dHL-60 migration than lysates ($p<0.007$). These results indicate that *T. tenax* is chemotactic for human neutrophils.

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Ecology of fear: ectoparasites exert non-consumptive effects on host larvae with consequences for adults.

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Hosts should be under strong selection to avoid contact with parasites or the infectious stages of parasites to minimize the risk of infection. Exposure to parasites has been shown to have deleterious effects on host fitness even in the absence of infection. The mere presence of parasites or parasite cues can influence host behavior, foraging habits, and space use – known cumulatively as non-consumptive effects (NCEs). However, it is unknown if exposure to parasites can impact larval development and whether these NCEs will have knock-on effects on the adult stage. We tested whether exposure to ectoparasitic mites during larval development affects *Drosophila nigrospiracula* pupation success and adult body condition. We found that larvae preferentially pupated away from mites in a choice-test and had reduced pupation rates overall. Moreover, the mean body mass of recently eclosed adult flies was significantly lower among exposed larvae compared to larvae that were not exposed to mites. Our results show that *Drosophila* larvae respond to mites despite mites not infecting the larvae. Parasite exposure during the larval stage elicited strong non-consumptive effects and these effects were transstadial, persisting across life stages. These indirect non-consumptive effects of parasite exposure represent an example of the “ecology of fear”, with significant consequences for host fitness and population growth. These findings suggest that the total impact of parasites on host populations and communities may have been previously underestimated.

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Decoding the molecular mechanisms of host manipulation by parasites.

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The water-seeking behavior exhibited by arthropod hosts infected with parasitic worms, including nematodes and nematomorphs, is a classic example of an extended phenotype. This aberrant host behavior facilitates the egression of mature worms into a favorable environment at the expense of the host. In this study, we investigated the molecular mechanisms underlying this behavior using the European earwig (*Forficula auricularia*) and its parasitic mermithid nematode (*Mermis nigrescens*) as a host-parasite model. To develop a genetic reference, we sequenced, assembled, and annotated the genomes of both organisms using Nanopore long-reads and 10x linked-reads. The earwig genome assembly resulted in 919 scaffolds, with the largest 20 scaffolds making up 50% of the genome. Similarly, the nematode genome assembly consists of 867 scaffolds and the largest 30 scaffolds make up 50% of the genome. We annotated 12,876 protein-coding genes and 21,031 mRNAs in earwigs and 12,313 protein-coding genes and 24,186 mRNAs in nematodes. Busco scores of over 90% suggest that both genomes are largely complete and high-quality. To understand the behavioral manipulation, we collected samples at three timepoints for both the host and the parasites, including early infection stage, late infection stage, and during manipulation stage, with samples from uninfected earwigs as

control. We conducted differential gene expression analysis with RNA-seq, and across all pairwise comparisons, 673 genes were upregulated and 593 genes were downregulated in earwig samples, while 2,672 genes were upregulated and 2,293 were downregulated in nematode samples. Cluster analysis and gene set enrichment analysis on the candidate genes showed transmembrane activity, signal transduction, serine endopeptidase, and cyclic nucleotides as common enriched pathways in earwigs and nematodes, suggesting increased intracellular communication within and between organisms. Pathways related to odorant binding were also significant in earwigs, indicating the role of environmental cues during the behavior manipulation process. Transmembrane signaling in response to environmental cues through cyclic nucleotide phosphodiesterase activity and odorant binding is a potential manipulation candidate pathway for further investigation. Our study provides valuable insights into the genomes, candidate pathways and genes, improving our understanding of the molecular regulation of water-seeking behavior in both the host and the parasite.

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To “drive” a landlubbing insect into deadly waters: a hairworm story.

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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. From subtle differences in host appearance to striking changes in host behavior, parasites can act upon a multitude of host biological systems. Even though this phenomenon was demonstrated empirically over 50 years ago, the causal chain between the manipulation factors expressed in a parasite and the observable changes in host phenotype remains elusive to this day. If the manipulation factors of a parasite are primarily expressed during key moments of its development, it is thus important to test for phenotypic host alterations throughout infection. 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 changes occur throughout hairworm development. We hypothesize that these modified behaviors 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 are testing multiple stimuli that may determine the extent to which hairworms can modulate host behavior, and whether these behaviors correlate with hairworm development. Here, we present preliminary data from these experiments and attempt to describe the sensory landscape governing positive hydrotaxis in infected crickets.

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Infected crickets eat less and convert food to energy less efficiently than uninfected crickets.

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Growth and reproduction are affected by protein and carbohydrate intake in the diet of insects. For female insects, whose fat body is the precursor for egg production, levels of carbohydrates are imperative for reproduction efforts. When insects are infected with parasites however, food resources are often sequestered by the developing parasites which has consequences to host fecundity. Hairworms are parasites of crickets and other arthropods that develop within the hemocoel of the host, consuming the fat as it develops. While great strides have been made in hairworm ecology and taxonomy in recent years, little research has been done on the impacts hairworms have on their host's development. In this study we tested the effect of parasitism on host diet choice and development of female house crickets (*Acheta domesticus*). Two diets were provided, one high in carbohydrates and one high in protein. We measured the food consumed and cricket preference for carbohydrates vs. proteins. We simultaneously examined the effects of their diet choice on host growth, fecal output, egg production, and lipid content. In addition, common nutritional indices were used to examine feeding efficiency and resource allocation in hosts infected with *Paragordious varius*. Compared to uninfected controls, infected crickets consumed the same proportions of protein to carbohydrates but consumed 14% less food than the uninfected controls despite parasites. We found a higher standard growth rate (SGR) in the control group compared to infected crickets but found a relationship between higher intensities of infection and lower SGR in infected crickets. Additionally, we found infected crickets had produced no eggs during the infection and had significantly lower lipids, ovipositor growth, and fecal output compared to controls, suggesting consequences to host fecundity. When using indices based on wet mass that incorporate the mass of parasites, there was no difference in converting intake to wet body mass. However, when using indices that incorporate dry mass (no parasites) to resource allocation, we found controls converted food intake 3.7× more efficiently than infected crickets. These results suggest that infected crickets assimilate energy less efficiently than controls because food intake is allocated to the developing hairworm.

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The size and body composition of mosquitoes is altered by environmental temperature and aging.

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The body temperature of mosquitoes fluctuates with the environmental temperature, and changes in body temperature alter a myriad of physiological processes. Moreover, as mosquitoes age, they senesce, which means that their physiology deteriorates over time. We hypothesized that as the temperature warms, the rate of senescence increases, and began to test this hypothesis by scrutinizing whether senescence-based deterioration is altered by environmental temperature. We reared mosquitoes at three different temperatures (27°C, 30°C and 32°C) and measured mosquito size and body composition at four different adult ages (1, 5, 10, and 15 days post-eclosion). Experiments uncovered that mosquitoes reared in warmer temperatures are smaller, but no significant changes occur in their carbohydrate or lipid content. Aging alters body size, as well as carbohydrate and lipid content, in ways that correlate with the recovery from metamorphosis and the ensuing senescence. Finally, protein content decreases with aging and at warmer temperature, and the aging-associated decrease occurs in younger individuals when the temperature is warmer. Overall, temperature and aging are two factors that individually and interactively alter the size and composition of adult mosquitoes.

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Embryonic female-lethal New World screwworm strains, high expression, low performance.

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New World screwworm, *Cochliomyia hominivorax*, is an economically important ectoparasite of warm-blooded animals in South America and the Caribbean. It primarily effects livestock but can infest humans. Efforts to control *C. hominivorax* led to the development of the sterile insect technique (SIT) during the mid-twentieth century. Recent efforts have been aimed at producing male-only strains, which would reduce costs and/or increase production. No methods to produce male-only releases have been developed or utilized by the eradication program. Female-lethal transgenic strains have been developed for screwworm but producing strains with the performance and reliability needed for mass-rearing has been challenging. Limiting expression of female-lethal transgenes to the early embryo is desirable since female lethality in later stages would increase the cost of feeding and could cause fouling of the diet. For use in a two-component tetracycline-off system, two strains with driver constructs designed with promoters of genes with high activity exclusively in embryos were developed. Driver construct DR7 utilized the promoter for *Nullo*, while driver DR6 utilized the promoter of an uncharacterized gene adjacent to *Nullo*. These driver strains were crossed to female lethal effector strains and evaluated for strain performance. Interestingly, DR6 strains caused pupal female mortality, while DR7 strains were lethal in embryo and early larval development when doxycycline is withheld. Comparison of the expression of tTA to biological performance suggests that strains with higher expression of tTA reduced fitness. Although neither promoter produced strains that meet the standards necessary for mass-rearing, these strains provided important insight into developing sex-lethal transgenic strains. Such as, selecting promoters with excessive expression may result in poor fitness and designing constructs with low or moderate expression may produce overall stronger strains.

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Testing behavior alteration in crickets experimentally infected with horsehair worms.

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Some parasites can alter host behavior to facilitate their transmission. However, in many instances of parasite manipulation, rigorous controlled studies demonstrating direct causation are lacking. Previous studies often use field-captured hosts that were naturally infected in the wild. Thus, they can only identify correlations between parasitic infection and behavior and fail to control for the alternative possibility that hosts exhibiting particular behaviors may be more likely to become parasitized. Studies lacking randomly assigned healthy control and experimentally-parasitized treatment groups often fail to distinguish among two alternative hypotheses for alteration in host behavior during infection – host-initiated defense against parasite or immune by-product of infection. Previous studies of the horsehair worm, *Paragordius varius*, suggest that, upon maturation in 28 days, *P. varius* manipulates its host into entering water where the parasite emerges, mates, and completes its life cycle. Yet, these studies failed to demonstrate causation due to the reliance upon observations of field-collected infected and uninfected crickets. Here, we experimentally or sham infect lab-raised crickets (*Acheta domesticus* and *Gryllus firmus*) with *P. varius* and compare their behavior to test the hypothesis that infection with the horsehair worm causes the following behavioral changes: 1) increased activity level, 2) increased phototaxis, and 3) increased tendency to jump into open water. We use EthoVision software to analyze

their behavior at weeks 1, 3, and 4 post-infection. Results will be discussed in the context of testing parasite manipulation in experimentally infected hosts.

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How surreal are the Escherbothriidae? A molecular perspective.

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The Escherbothriidae is currently a small cestode family in the order Rhinebothriidea with 14 species in the genera *Escherbothrium* and *Stillabothrium*. Escherbothriids are distinguished from other rhinebothriideans by possessing unusual scolex morphologies. While the bothridia of most rhinebothriideans possess multiple rows of loculi, those of species of *Stillabothrium* possess loculi arranged in rows and columns. Even more unique are the bothridia of *Escherbothrium* with diagonal septa delimiting triangular loculi. There are a few other rhinebothriidean taxa that also exhibit unusual bothridial morphologies whose familial affinities remain in question. Species of *Scalithrium*—a genus currently assigned to the Rhinebothriidae that includes species hosted by rays and skates—unlike all other rhinebothriideans, have bothridia divided into multiple rows of loculi but lack a longitudinal septum. Species of *Rhinebothrium* in skates also lack this longitudinal septum, raising doubt about their generic placement. Also parasitizing skates are species in the genus *Semiorbiceptum*, whose bothridia not only lack a longitudinal septum, but also exhibit semicircular septa in their central region. Considering the challenges of assigning these taxa to family based on morphology alone, the main goal of this study was to assess their phylogenetic position using molecular data. Partial 28S rDNA sequence data were generated for eight rhinebothriidean species, four of which are undescribed. These data, along with those for representatives of all five rhinebothriidean families, were included in a Maximum Likelihood phylogenetic analysis. Additionally, morphological data were collected using light and scanning electron microscopy for three of the four undescribed species. The resulting phylogeny suggests that the Escherbothriidae is much more diverse than initially thought. The family includes subclades of *Stillabothrium* and *Escherbothrium*, and a subclade consisting of all *Scalithrium* and *Semiorbiceptum* species, as well as the species assigned to *Rhinebothrium* that parasitize skates. Recognition of a skate-hosted subclade within the family and transfer of relevant species to *Semiorbiceptum* resolves the non-monophyly of *Scalithrium* and *Rhinebothrium*. In combination with the description of a new species of *Escherbothrium* and two new species of *Semiorbiceptum*, these taxonomic actions bring the total number of species in the Escherbothriidae to 33. Supported by NSF grants 1921404 & 1921411.

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Naming cryptic parasite species with molecular morphology.

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Due to morphological convergences and trait reductions, there are numerous cryptic parasite species. For instance, Apicomplexa — the parasitic protist phylum that includes malaria pathogens — may have millions of species, but only a few thousand have been named. While this example is extreme in scale, many parasite groups face the exact same issue. Without names, it is difficult to communicate about species, which can slow the rate of scientific progress. Traditionally, morphology of visualizable features has been key to naming and describing species, but clearly is insufficient for naming many parasite groups. To help alleviate this problem, we propose that “molecular morphology” can be used to name species, as molecules very much have physical morphological traits. For instance, proteins can be sequenced and modeled with computers or, if necessary, visualized with X-ray crystallography. There are typically thousands of proteins per species, and they tend to vary in terms of structure, function, and evolutionary rates, just as traditional morphology does. Furthermore, using genomic sequencing, the presence or absence of protein coding genes makes a simple binary character set (called gene content) that can easily distinguish species as well as broader lineages. We believe molecular morphology is in principle no different than traditional morphology and accordingly it is easily integrated into current best practices for using Linnaean taxonomy. With molecular morphology, parasites can be named more expediently and accurately.

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The more you know the better the estimate.

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Generating an accurate estimate of the full extent of the diversity of life on the planet is one of the most intriguing, yet challenging goals of biodiversity research. Such efforts typically involve combining estimates generated separately for different taxonomic groups. Many approaches for making such estimates have been proposed. Some draw on macroecological patterns (e.g., latitudinal gradients, species-area relationships) others are based on taxonomic effort (e.g., time-species accumulation curves, authors-species accumulation curves), yet others use diversity ratios (e.g., ratios between taxa, host-specificity and spatial ratios). However, the limitations of all such approaches abound. We argue that accurate global estimates of parasite diversity are much more achievable than estimates of free-living taxa using diversity ratios because of two fundamental features of parasite biology. The first is that parasites are, by definition, associated with hosts. The second is that all parasites exhibit some degree of host specificity. Units of assessment thus exhibit clearly defined boundaries and estimates are easily replicated. Especially tractable are parasites of vertebrates because vertebrate diversity is extremely well documented and vertebrate classification schemes tend to reflect phylogenetic relationships. Obviously, the simplest cases are parasites that exhibit oioxenous specificity. We used this approach in 2017 to estimate the global diversity of 15 of the 16 orders of tapeworms that parasitize elasmobranchs. At that time, the breadth, but not necessarily the depth, of sampling across elasmobranch taxa was already fairly extensive. Existing cestode records from host orders, families, genera, and species were used to inform our predictions of the full spectrum of actual host taxa and thus also cestode species in each group. We predicted that the total number of 729 described cestode species was an order of magnitude lower than the actual global total. Since then, the cestodology community has described 195 new species of elasmobranch cestodes. All but 13 of these species parasitize elasmobranchs in genera we predicted would host cestodes of the relevant order and thus are consistent with our global predictions. In five cases, the number of cestode species parasitizing host genera now exceeds the number we predicted by one to five species.

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A new genus and species of Cathariotrematinae Bullard, 2021 (Monogenoidea: Monocotylidae) infecting the olfactory lamellae of deepwater gulper sharks, *Centrophorus granulosus* (Squaliformes: Centrophoridae) in the Gulf of Mexico.

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Species of Monocotylidae comprise ectoparasites that collectively infect the gill, olfactory bulb, urogenital system, and body surface of sharks, rays, and chimaeras (Chondrichthyes). The recently proposed monocotylid subfamily Cathariotrematinae includes 5 genera comprised wholly of species infecting the olfactory bulb of sharks and a chimaera only. During a recent parasitological survey of epipelagic and deepwater fishes in the Gulf of Mexico, we longlined several specimens of the deepwater gulper shark, *Centrophorus granulosus* (Squaliformes: Centrophoridae) and examined them for monocotylid infections. Despite being commonly caught on deepwater longlines, relatively little is known about the parasites that infect this shark and no record of a monocotylid from this host was previously known. Upon examining these sharks, we collected several monocotylid specimens infecting the olfactory bulb. These monocotylids were fixed in 10% formalin for morphology and preserved in 95% EtOH for DNA extraction and subsequent phylogenetic analysis. The specimens comprise a new cathariotrematine genus by having a haptor comprising a central loculus (without paired depressions), 5 peripheral loculi, a tripartite attachment organ comprising the posterior-most 3 loculi, and small hamuli. This is only the second species of the subfamily documented from the Gulf of Mexico. A phylogenetic analysis based on the large subunit ribosomal DNA (28S) recovered a monophyletic Cathariotrematinae and the new species sister to monotypic Triloculotrema.

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The Mongolian Remodeling and the structure of host-parasite diversity across Central Asia.

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At the boundary between the Eocene and Oligocene epochs, Central Asian landscapes underwent a dramatic change due to climatic cooling and aridification. Dense forests gave way to open grasslands and large-bodied mammal populations declined while small rodents diversified, a faunal turnover known as the Mongolian Remodeling. Throughout the subsequent Oligocene and Miocene epochs, the landscapes of Central Asia endured repeating ecological disruptions due to climatic oscillation. These disruptions drove species diversification and structured complex host-parasite communities. Rodents in Central Asia host a diverse array of endoparasite species, providing an opportunity to evaluate the consequences of the Mongolian Remodeling for host-parasite communities. In this study, I quantify the diversity and evaluate the biogeographic history of a hyper-diverse group of cestodes, the *Paranoplocephala* species complex (family: Anoplocephalidae), to determine the temporal and geographic origins of this group. Using a large series of anoplocephalid cestode samples collected from Mongolia, I sequenced the mitochondrial COI gene from 220 individual worms. These sequences were compared to a reference database of specimens of confirmed species identity to determine the scope of diversity represented in my dataset. Based on these results, I sequenced whole mitochondrial genomes of representative samples to build a more robust phylogeny than has been possible based on COI alone. I have revealed eleven undocumented genetic lineages that may represent undescribed species warranting further taxonomic investigation. Biogeographic reconstructions are consistent with the hypothesis that the *Paranoplocephala* species complex originated in Central Asia shortly after the Mongolian Remodeling, approximately 30 million years ago, followed by an extended period of diversification. These results suggest that the repeating ecological disruption and climatic changes that occurred throughout the Oligocene epoch in Central Asia played an important role in structuring the diversity of the *Paranoplocephala* species complex.

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Taxonomy, systematics, and new keys for *Caligus* (Copepoda: Caligidae) illuminated by confocal laser scanning microscopy.

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Sea lice, members of the copepod family Caligidae, are the most economically important parasitic copepods and some of the most common parasites on marine fish. The Caligidae is the most species-rich copepod family with 516 species in 28 genera and *Caligus* is the most species-rich genus in the whole of the Copepoda, with 276 described species. Because of the prominence of these fish parasites in natural ecosystems and their burgeoning economic importance as pathogens in aquaculture, it is important to be able to identify these parasites in a timely manner. However, identifying these taxa, especially navigating the 276 species in *Caligus*, is a major hurdle. In this study, we use confocal laser scanning microscopy (CLSM) to provide high resolution, 3D images of caligid copepods and score all valid species of *Caligus* for 41 morphological characters, the results of which inform their taxonomy and systematics. To facilitate the identification of caligids, we have developed three keys including a key to the caligiform families, a key to the genera of Caligidae, and a key to the species of *Caligus*. A dichotomous key to the 276 species of *Caligus* would be lengthy, difficult to navigate, and rapidly outdated given that several new species are typically described each year. To overcome these limitations, we are developing an interactive key to *Caligus* based on a character matrix comprising 41 morphological characters and several additional body proportions scored for all valid species of *Caligus*. The key will be presented an easy-to-navigate online format with accompanying CLSM images for each character state in the matrix.

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Implant device that enables in vivo study of helminths and mimics parasite antigen load resulting in a novel vaccine strategy.

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Many parasites develop a relationship with the host whereby maintenance of a chronic low level infection induces a host response that protects against high numbers of parasites and clinical disease. Our hypothesis is that an implantable device can maintain a low level of parasites in the host, mimicking this phenomenon and inducing protective immunity. Herein, we demonstrate the utility of the implant device for protection of sheep against *Haemonchus contortus*. In a proof-of-concept study, sheep were vaccinated using a soluble preparation or implant containing parasite antigens. Challenge studies revealed the implant was effective in decreasing the number of adult worms present. In subsequent experiments, live *Toxocara canis* larvae were maintained in the device without causing neural larva migrans in mice and enabling recovery of L3 without laborious dissection of individual granulomas. Thus, the device can be used to maintain parasites and parasite antigens within vertebrate hosts without harming the host directly. Moreover, live parasites can be recovered without having to sacrifice the

host, which will make studies in natural hosts more practical. Future studies will target refinement of antigen release and strategies to capture cells and molecules involved with parasite/host interaction.

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Characterization and process development of serine protease inhibitor (Sj-B6): A next generation transmission-blocking veterinary mRNA vaccine for asiatic schistosomiasis.

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Asiatic schistosomiasis caused by *Schistosoma japonicum* is a neglected tropical disease resulting in significant morbidity to both humans and animals - particularly bovines - in endemic areas. Infection with this parasite leads to less healthy herds, causing problems in communities which rely on bovines for farming, milk and meat production. Additionally, excretion of parasite eggs in feces perpetuates the life cycle and can lead to human infection. We endeavored to develop an inexpensive and effective mRNA vaccine based on secretory serine protease inhibitor (serpin) from *S. japonicum* (Sj-B6). In pathogens, serpins are believed to have evolved specifically to limit host immune responses by interfering with the host immune-stimulatory signals. Transcriptional profiling and proteomics demonstrated that Sj-B6 is expressed in the intra-mammalian life cycle stages but particularly in the eggs, suggesting a possible role in disease transmission. Recombinant Sj-B6 inhibited host pancreatic elastase in a dose-dependent manner and was strongly recognized by experimentally infected rat (naturally-resistant hosts) sera when compared to chronically-infected mouse counterparts, indicating that rSj-B6 is not only highly immunogenic, but critically involved in disease resistance. This study presents a comprehensive functional characterization of Sj-B6 supporting its further development as a vaccine candidate against Asiatic schistosomiasis. A pilot study evaluating the efficacy of a novel Sj-B6 mRNA vaccine using a proprietary technology (HDT-301) invented by our partners at HDT Bio is now underway. The HDT-301 platform consists of a self-replicating RNA (repRNA) adsorbed and stabilized on a Lipid InOrganic Nanoparticle (LION™) carrier. The repRNA/ LION™ vaccine stabilizes the RNA in vivo allowing it to persist longer. A successful veterinary vaccine would play a major role in reducing pathogen transmission to humans by interrupting the parasite life cycle and improving quality of life for people living in endemic countries.

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Kiss and spit metabolomics highlights the role of host purine metabolism during pathogen infection.

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Intracellular bacteria and protists are auxotrophic for many metabolites and must rely on the host cell to supply these nutrients. The mechanisms of how pathogens manipulate host metabolism to their benefit are not totally understood. These questions are difficult to address for intracellular pathogens because one cannot easily distinguish the origin of the metabolite as host or pathogen derived. The obligate intracellular parasite *Toxoplasma gondii* manipulates the host cell by a pre-invasion process called “kiss

and spit", where the contents of the parasite rhoptry organelles are secreted into the host cytoplasm before invasion occurs. We used metabolomics, genetic and chemical inhibition and click chemistry to analysis the *T. gondii* kiss and spit effect on host cell metabolism. We found that rhoptry contents from kiss and spit altered metabolite abundance in nucleotide synthesis, the pentose phosphate pathway, glycolysis, and amino acid synthesis. An increase in 2,3-bisphosphoglycerate (2,3-BPG) abundance led us to investigate the activation of host cytosolic nucleosidase II (cN-II) to provide purines for the parasite.

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Detection of trypanosomatid parasites in bee assassin bugs (*Apiomerus* spp.) in Keith County, Nebraska.

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Trypanosomatids are a diverse group of flagellates primarily infecting insects, vertebrates, and plants. Among them, dixenous species such as species of *Trypanosoma* have been studied intensively because of their medical and economic importance; however, monoxenous species are common in insects, and remained mostly neglected. These species undergo life cycles with evident low levels of host-specificity, high probability of nonspecific infection, and some genetic and physiological adaptations that could increase the chances of establishing in a new host. Furthermore, some of these trypanosomes could harbor viruses. The objective of this work was to assess the presence of trypanosomatids in the bee assassin bug *Apiomerus* spp. (Hemiptera: Reduviidae) in a location near the sandhills, in west-central Nebraska. Individuals were collected manually from plants where they were feeding on other insects. Also, some potential prey items were collected. Digestive tract of the *Apiomerus* was removed to extract the intestinal contents and these were smeared onto slide. Samples were stained with Giemsa and examined with optical microscopy. Some individuals were maintained alive for assessing feeding habits. One hundred percent prevalence of flagellated forms was found in the digestive tract of *Apiomerus* spp. (n=35); trypomastigotes were also found in both saliva and feces. Some flagellated structures were found in potential prey species (dipterans and hymenopterans). Cannibalism habits in *Apiomerus* were recorded when the food supply was absent. Discovery of trypanosomatids in these bugs and perhaps in their prey species suggests an underestimated diversity of flagellated parasites in the central Nearctic, given the relatively low number of insect species examined so far. Moreover, the study of the relationships of these parasites with their insect hosts could give us a more complete understanding of the parasite dynamics of pathogenic and non-pathogenic trypanosomatid species, subsequently establishing monitoring processes in the frame of emerging diseases identification and potential prevention.

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Volatiles in 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 (*Tenebrio* sp. or *Tribolium* sp.) than feces

from uninfected rats. One study showed that a volatile chemical (or chemicals) in infected rat feces attracts the intermediate host *Tribolium confusum*, though the volatile chemical(s) have never been identified. The aim of this study was to determine the volatiles in feces of rats infected with *H. diminuta*. Four 8-week-old female Sprague Dawley rats were infected with *H. diminuta* (Virginia Tech IACUC protocol 22-050). Fecal samples pre- and post-infection were collected. Five carboxylic acid volatiles, identified with GC-MS analysis, were isolated only from post-infection 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. Experiments to demonstrate the beetle attraction to the volatiles are underway. This is the first report identifying the volatiles found in feces of rats infected with *H. diminuta*.

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A review of *Pfeifferinella* (Apicomplexa: Pfeifferinellidae) infections in North American freshwater gastropods.

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Coccidia of the genus *Pfeifferinella* Wasielewski, 1904, infect and develop in the digestive gland of snails, and oocysts enter the digestive track and are released in the feces. The oocysts are characterized as octozoic, with a large oocyst residuum, without sporocysts, and a distinct convex micropyle usually present at one end of the oocyst. The genus contains four described and at least 1 undescribed species infecting European, North American, and North African Freshwater and terrestrial gastropods. However, as a group, the coccidia of pulmonate mollusks have been poorly studied, and relatively little is known of their basic biology. This is particularly true for North America as only one publication reports *Pfeiferinella* species infecting freshwater gastropods, including *Stagnicola elodes* (Lymnaeidae), *Physa gyrina* and *Aplexa hypnorum* (Physidae) and *Planorbella trivolvis* (Planorbidae) and collected from Minnesota and Washington. To evaluate how commonly freshwater snails are infected with these coccidia, we sampled seven species of freshwater snails from multiple locations in Arkansas, Oklahoma and Wisconsin, and examined their feces for coccidian oocysts. We document octozoic oocysts with a large oocyst residuum which lacked sporocysts from *Planorbella* c.f. *trivolvis* and snails in the genus *Physa* collected from Arkansas and octozoic oocysts with a large oocyst residuum which lacked sporocysts of a second morphotype from *Planorbella* c.f. *trivolvis* snails collected from Wisconsin. Our study presents the largest dataset on coccidia of freshwater snails to date and a number of hypotheses for additional research, including questions regarding the variation of coccidia host specificity and virulence will be discussed.

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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
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