PROCEEDINGS

AAVP

AMERICAN ASSOCIATION OF VETERINARY PARASITOLOGISTS

51st Annual Meeting
July 15-18, 2006

and

MEMBERSHIP DIRECTORY

AMERICAN ASSOCIATION OF VETERINARY PARASITOLOGISTS AWARDS HISTORY

Distinguished Veterinary Parasitologists

1985	Jitender P. Dubey
1986	Norman D. Levine
1987	E. J. Lawson Soulsby
1988	Jeffrey F. Williams
1989	K. Darwin Murrell
1990	William C. Campbell
1991	Jay Hal Drudge and Eugene T. Lyons
1992	Gilbert F. Otto
1993	Thomas R. Klei
1994	Peter M. Schantz
1995	James C. Williams
1996	T. Bonner Stewart
1997	J. Owen D. Slocombe
1998	J. Ralph Lichtenfels
1999	Roger K. Prichard
2000	Edward L. Roberson
2001	Byron L. Blagburn
2002	Sidney A. Ewing
2003	Louis C. Gasbarre
2004	David S. Lindsay
2005	Jorge Guerrero

2006 Distinguished Veterinary Parasitologist Award Winner

JOHN W. McCALL, Ph.D.

Biographical Sketch

JOHN W. McCALL

John W. McCall was born in Blackshear, Georgia, in 1941 and grew up in Alma, another very small town nearby. He was fortunate to work with Mr. Frank E. Johnson, a local pharmacist who was his sixth grade teacher's husband, from the age of 12 until he left home to attend the University of Georgia (UGA) in Athens as a freshman in 1959. Upon receiving a BS Degree in Zoology, he was introduced to the field of parasitology by Dr. R. Barclay McGhee, who encouraged him to do his graduate work in Medical Entomology under Dr. A.B. Weathersby. Dr. Weathersby later became a close personal friend, as well as a mentor. He received his MS (1967) and PhD (1970) Degrees from UGA while working under a Malariology Training Program, which was funded by the NIH and co-directed by Drs. McGhee and Weathersby. In 1970, he moved across the campus to join the faculty of the College of Veterinary Medicine as a Research Associate in Dr. Paul E. Thompson's filariasis laboratory, where he worked with several colleagues (Drs. Tom Klei, Jack Malone, Hyong-Sun Ah and Macki Donahoe)—a couple of whom are well-known to the AAVP. He was promoted to Associate Professor in the Department of Parasitology in 1978 and to Professor in 1983. He remained with the College through several departmental name changes (now, Department of Infectious Diseases) until his retirement to Professor Emeritus on 01 May 2006. During his academic career, he contributed to the teaching of veterinary parasitology to veterinary students and graduate students with various colleagues (Drs. Ed Roberson, Bill Hanson, Katherine Prestwood, Ray Kaplan, Susan Little, David Peterson), supervised Research Associates (Drs. Mike Dzimianski, Prasit Supakorndej, Lari Cowgill, Bruce Lindemann, Jacqueline Testa) and visiting scholars (Drs. Luigi Venco, Barbara Di Sacco, Leucio Alves), directed programs of study for five Master's Degree and seven PhD Degree students, served as Director of the NIH-Sponsored Filariasis Repository Service (now, the Filariasis Research Reagent Resource Center) for 35 years and sustained a filariasis research program funded mainly by the NIH and WHO thoughout most of his academic career.

Dr. McCall is currently Vice President of the American Heartworm Society and a charter member of the American Heartworm Society, the Merial-sponsored International Feline Heartworm Disease Council and the Pfizer-sponsored Global Advisory Council on Diseases of Small Animals. He served on the International Committee for Developing Guidelines for Evaluating Anthelmintics for Small Animals for the World Association for the Advancement of Veterinary Parasitology, and as Reviewer for the NIH Delta Primate Research Center (Tulane University), the NIH ICIDR Program Grants, the NIH Tropical Disease Research Center Program Grants, and the NIH Small Business Grants Programs. He was an invited speaker for the U.S.-Japan Cooperative Medical Science Program almost annually from 1971 to 1978 and was on numerous World Health Organization (WHO) Scientific Working Groups on Filariasis almost annually from 1977 to 2001. He has held Visiting Professorships in Lima, Peru; Milan, Italy; Rio de Janeiro, Brazil; and Recife, Brazil and has been an AID/NIH consultant to The Hebrew University in Jerusalem and the Ain Shams University in Cairo. He has published 157 original research articles in peer-reviewed journals, 40 non-referred scholarly articles and reports, and 11 book chapters. He has been an invited speaker for meetings of the American Veterinary Medical Association, North American Veterinary Conference, American Heartworm Society, World

Association for the Advancement of Veterinary Parasitology, European Multicolloqium of Parasitology, World Small Animal Veterinary Association World Conference, Western Veterinary Conference, Southwestern Veterinary Conference, Southeastern Veterinary Conference, Atlantic Veterinary Conference, and American Academy of Veterinary Pharmacology and Therapeutics. He has presented over 285 invited lectures at national or international meetings and continuing education seminars for veterinarians and presented or co-authored over 210 additional scientific papers. He was president of the Southeastern Society of Parasitologists from 1978 to 1979 and has been President of TRS Labs, Inc., his privately owned contract research laboratory, since 1980. Although officially retired from UGA, he continues to be actively engaged in research at both UGA and TRS Labs.

John and his wife Gail (also sweetheart and best friend) have been married for the past 42 years. They have two children, Christy McCall and Sharon Carter, and four grandchildren (Austin, Ashley, Evan and Savannah). Scott McCall and Justin Carter are not only great sons-in-law and friends, but also contribute greatly to the day-to-day operation of TRS Labs.

AMERICAN ASSOCIATION OF VETERINARY PARASITOLOGISTS AWARDS HISTORY

Hoechst-Roussel Agri-Vet Company Graduate Student Research Award

1987	Lora G. Rickard
1988	Debra A. Cross
1989	Stephan C. Barr
1990	Jeff C. Parsons
1991	Carlos E. Lanusse
1992	David G. Baker
1993	Rebecca A. Cole
1994	Ray M. Kaplan
1995	Steve T. Storandt
1996	A. Lee Willingham III
1997	Carla C. Siefker
1998	Ryan M. O'Handley
1999	Jon S. Matthew
2000*	Sheila Abner
2001	Andrew Cheadle
2002	No recipient
2003	Mary G. Rossano
2004	Andrea S. Varela
2005	Alexa C. Rosypal
2006	Sheila Mitchell

*As of 2000, this award was renamed the "Intervet Graduate Student Research Award".

Distinguished Service Award

1976	Rurel R. Bell
1983	Terrance J. Hayes
1987	Norman F. Baker
1988	Donald E. Cooperrider
1994	S. D. "Bud" Folz
1997	Honorico Rick Ciordia

2006 Distinguished Service Award Winner

RAFFAELE RONCALLI AMICI, D.V.M., M.S.

Biographical Sketch

RAFFAELE RONCALLI AMICI

Raffaele Roncalli Amici, a native of Italy, obtained his D.V.M. degree from the University of Perugia, Italy in 1949. After several years as a researcher at the Veterinary School, University of Milan, he was awarded a Fulbright Scholarship to attend the University of Georgia, where he received a M.S. degree working under Dr. W. Malcolm Reid (1960). In ensuing years he held several managerial positions with Hoffmann-La Roche, Inc and Merck & Co., Inc. in the USA; with the latter company he was in charge of international veterinary research. In this capacity, for many years he conducted clinical trials for the development of new veterinary products (i.e. anthelmintics such as thiabendazole, rafoxanide and ivermectin and coccidiostats such as amprolium and nicarbazin) in Europe, Latin America, Australia and Japan. He is a Past-President of the New Jersey Society for Parasitology (1972-1973) and a former Chairman of the International Section, Animal Health Institute, Washington, D.C. He has been a visiting professor (veterinary parasitologist) at the University of Naples (1985, 1992, 2002, 2003, 2004) and at the University of Bari (1991). He is the author of more than 100 scientific publications, especially in the field of parasitology and the history of parasitology. Dr. Roncalli spent a number of years in Japan; he is a member of the Japanese Veterinary History Society and has lectured extensively on the history of Japanese veterinary medicine; also, he was President of the Ukiyo-e Society of America (1985-1987). Dr. Roncalli twice has been President of the American Veterinary Medical History Society (1988-1989; 2004-2005); presently he is the historian of the American Association of Veterinary Parasitologist.

AMERICAN ASSOCIATION OF VETERINARY PARASITOLOGISTS
Founded 1956

Officers 2005-2006

President	Ann R. Donoghue PR Pharmaceuticals, Inc. Fort Collins, CO
President-Elect	Daniel E. Snyder Elanco Animal Health Greenfield, IN
Vice-President	David S. Lindsay Virginia Tech Blacksburg, VA
Secretary/Treasurer	Alan A. Marchiondo IVX Animal Health, Inc. St. Joseph, MO
Immediate Past-President	Linda S. Mansfield Michigan State University East Lansing, MI

2005-2006 AAVP Committee Chairs

Program – David S. Lindsay, (Chair) and Sheila M. Mitchell, Blacksburg, VA
Archives – Tom Nolan, Philadelphia, PA
Historian - Raf Roncalli, Milltown, NJ
Awards - Sara Marley, Duluth, GA
Constitution/Bylaws - Gil Myers, Magnolia, KY
Education - Dwight Bowman, Ithaca, NY
Finance - Larry Cruthers, Corapeake, NC
Newsletter - Patrick Meeus, Kalamazoo, MI
Nominations – Pat Payne, Manhattan, KS
Outreach/Research - David Granstrom, Beltsville, MD
Publications/Internet - Doug Colwell, Lethbridge, Alberta, Canada
Student Representative - Isabelle Verzberger, Prince Edward Island, Canada
List Serve Manager - Bert Stromberg, St. Paul, MN

PAST PRESIDENTS OF THE AMERICAN ASSOCIATION OF VETERINARY PARASITOLOGISTS

1956-1958	Leonard E. Swanson
1958-1960	Fleetwood R. Koutz
1960-1962	William H. Krull
1962-1964	Saeed M. Gaafar
1964-1966	Everett D. Besch
1966-1968	George C. Shelton
1968-1970	John H. Greve
1970-1972	Harold J. Griffiths
1972-1973	Donald E. Cooperrider
1973-1975	Demetrice L. Lyles
1975-1977	Harold J. Smith
1977-1979	Norman F. Baker
1979-1981	Edward L. Roberson
1981-1983	Jeffrey F. Williams
1983-1985	John B. Malone
1985-1986	Robert M. Corwin
1986-1987	K. Darwin Murrell
1987-1988	Thomas R. Klei
1988-1989	Harold C. Gibbs
1989-1990	Bert E. Stromberg
1990-1991	Roger K. Prichard
1991-1992	J. Owen D. Slocombe
1992-1993	Ronald Fayer
1993-1994	George A. Conder
1994-1995	Charles H. Courtney
1995-1996	Byron L. Blagburn
1996-1997	Peter M. Schantz
1997-1998	James C. Williams
1998-1999	Louis C. Gasbarre
1999-2000	Robert S. Rew
2000-2001	Thomas J. Kennedy
2001-2002	Anne M. Zajac
2002-2003	Joseph F. Urban, Jr.
2003-2004	Craig R. Reinemeyer
2004-2005	Linda S. Mansfield
2005-2006	Ann R. Donoghue

PAST SECRETARY-TREASURERS OF THE AMERICAN ASSOCIATION OF VETERINARY PARASITOLOGISTS

William H. Krull	1956-1959
Edward G. Batte	1960
Donald E. Cooperrider	1961-1969
Rurel Roger Bell	1969-1977
Terence J. Hayes	1978-1983
Vassilios J. Theodorides	1983-1986
S.D. "Bud" Folz	1987-1992
Thomas J. Kennedy	1993-1998
Daniel E. Snyder	1998-2004
Alan A. Marchiondo	2004-

PAST AAVP ANNUAL MEETINGS

1956	1st Annual Meeting – SAN ANTONIO, TX 16 OCT
1957	2nd Annual Meeting – COLUMBUS, OH 17 AUG
1958	3rd Annual Meeting – PHILADELPHIA, PA 18 AUG
1959	4th Annual Meeting – KANSAS CITY, MO 23 AUG
1960	5th Annual Meeting – DENVER, CO 14 AUG
1961	6th Annual Meeting – WEST LAFAYETTE, IN 20 AUG
1962	7th Annual Meeting – MIAMI BEACH, FL 12 AUG
1963	8th Annual Meeting – NEW YORK CITY, NY 28 JUL
1964	9th Annual Meeting – CHICAGO, IL 19 JUL
1965	10th Annual Meeting – PORTLAND, OR 11 JUL
1966	11th Annual Meeting – LOUISVILLE, KY 13-14 JUL
1967	12th Annual Meeting – DALLAS, TX 9 JUL
1968	13th Annual Meeting – BOSTON, MA 21 JUL
1969	14th Annual Meeting – MINNEAPOLIS, MN 13 JUL
1970	15th Annual Meeting – LAS VEGAS, NV 22 JUN
1971	16th Annual Meeting – DETROIT, MI 18 JUL
1972	17th Annual Meeting – NEW ORLEANS, LA 17 JUL
1973	18th Annual Meeting – PHILADELPHIA, PA 15 JUL
1974	19th Annual Meeting – DENVER, CO 21 JUL
1975	20th Annual Meeting – ANAHEIM, CA 13 JUL
1976	21st Annual Meeting – CINCINNATI, OH 19 JUL
1977	22nd Annual Meeting – ATLANTA, GA 11 JUL
1978	23rd Annual Meeting – DALLAS, TX 17 JUL
1979	24th Annual Meeting – SEATTLE, WA 22-24 JUL
1980	25th Annual Meeting – WASHINGTON, D.C. 20-22 JUL
1981	26th Annual Meeting – ST. LOUIS, MO 19-20 JUL
1982	27th Annual Meeting – SALT LAKE CITY, UT 18-19 JUL
1983	28th Annual Meeting – NEW YORK, NY 17-18, JUL
1984	29th Annual Meeting – NEW ORLEANS, LA 15-17 JUL
1985	30th Annual Meeting – LAS VEGAS, NV 22-24 JUL

1986 31st Annual Meeting – ATLANTA, GA 20-22 JUL
1987 32nd Annual Meeting – CHICAGO, IL 19-21 JUL
1988 33rd Annual Meeting – PORTLAND, OR 17-18 JUL
1989 34th Annual Meeting – ORLANDO, FL 16-18 JUL
1990 35th Annual Meeting – SAN ANTONIO, TX 21-24 JUL
1991 36th Annual Meeting – SEATTLE, WA 28-30 JUL
1992 37th Annual Meeting – BOSTON, MA 2-4 AUG
1993 38th Annual Meeting – MINNEAPOLIS, MN 17-20 JUL
1994 39th Annual Meeting – SAN FRANCISCO, CA 9-12 JUL
1995 40th Annual Meeting – PITTSBURGH, PA 6-10 JUL
(Joint meeting with the American Society of Parasitologists)
1996 41st Annual Meeting – LOUISVILLE, KY 20-23 JUL
1997 42nd Annual Meeting – RENO, NV 19-22 JUL
1998 43rd Annual Meeting – BALTIMORE, MD 25-28 JUL
1999 44th Annual Meeting – NEW ORLEANS, LA 10-13 JUL
2000 45th Annual Meeting – SALT LAKE CITY, UT 22-25 JUL
2001 46th Annual Meeting – BOSTON, MA 14-17 JUL
2002 47th Annual Meeting – NASHVILLE, TN 13-16 JUL
2003 48th Annual Meeting – DENVER, CO 19-23 JUL
2004 49th Annual Meeting – PHILADELPHIA, PA 24-28 JUL
(Joint meeting with the American Society of Parasitologists)
2005 50th Annual Meeting – MINNEAPOLIS, MN 16-19 JUL
2006 51st Annual Meeting – HONOLULU, HI 15-18 JUL

AAVP 51st ANNUAL MEETING
Sheraton Waikiki Hotel

REGISTRATION

Ballroom Foyer - Second Level
Saturday, 15 July 2006, 12:00 Noon – 6:00 PM
Sunday, 16 July 2006, 8:00 AM – 5:00 PM
Monday, 17 July 2006, 8:00 AM – Noon

SPEAKER READY ROOM

Iao Needle/Akaka Falls Room – Second Level
Saturday, 15 July 2006, 1:00 PM – 6:00 PM
Sunday, 16 July 2006, 8:00 AM – 6:00 PM
Monday, 17 July 2006, 8:00 AM – 6:00 PM

SOCIAL PROGRAM

Welcoming Reception – Bayer Health Care, Animal Health
Moloka'i Ballroom – Second Level
Saturday, 15 July 2006, 7:00 PM – 9:00 PM

Merial Ltd Social
Moloka'i Ballroom – Second Level
Sunday, 16 July 2006, 6:30 PM – 9:30 PM

Fort Dodge Animal Health Social
Moloka'i Ballroom – Second Level
Monday, 17 July 2006, 6:30 PM – 9:00 PM

AAVP Graduate Student/Post Doc Luncheon/Mixer
Hilo Meeting Room – Second Level
Sunday, 16 July 2006, Noon – 2:00 PM

PRESIDENT'S AVMA/AAVP JOINT SYMPOSIUM

Hawai'i Convention Center - Room 309
Tuesday, 18 July 2006, 8:30 AM – 10:00 AM

AMERICAN ASSOCIATION OF VETERINARY PARASITOLOGISTS 51ST ANNUAL MEETING SPONSORS

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CORPORATE SPONSORS

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[1]AAVP Best Student Paper Presentation Awards
[2]Honorarium for Distinguished Veterinary Parasitologist Award
[3]Honorarium for AAVP Graduate Student Award
[4]In-Kind Meeting and Annual Association Support

GENERAL MEETING SPONSORS

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The American Association of Veterinary Parasitologists (AAVP) gratefully acknowledges the above companies for their support and sponsorship of the 2006 annual meeting.

American Association of Veterinary Parasitologist
51st Annual Meeting
Sheraton Waikiki Hotel, Honolulu, HI

SATURDAY JULY 15	SUNDAY JULY 16		MONDAY JULY 17		TUESDAY JULY 18
IAO NEEDLE/AKAKA FALLS ROOM SECOND LEVEL SPEAKER READY ROOM 1:00 PM - 6:00 PM	**IAO NEEDLE/AKAKA FALLS ROOM SECOND LEVEL** SPEAKER READY ROOM 8:00 AM - 6:00 PM		**IAO NEEDLE/AKAKA FALLS ROOM SECOND LEVEL** SPEAKER READY ROOM 8:00 AM - 6:00 PM		**CONVENTION CENTER** AVMA SPEAKER READY ROOM 7:00 AM - NOON
FORUM FOYER-OUTSIDE BALLROOM AAVP REGISTRATION TABLE 12:00 AM - 6:00 PM	**FORUM FOYER-OUTSIDE BALLROOM** AAVP REGISTRATION TABLE 8:00 AM - 5:00 PM		**FORUM FOYER-OUTSIDE BALLROOM** AAVP REGISTRATION TABLE 8:00 AM – 12:00 PM		
KOKO CRATER ROOM AAVP EXEC. CMTE. Board Meeting/Lunch 8:00 AM – 1:00 PM **KAUAI BALLROOM** OPENING REMARKS: President Ann Donoghue VP/Program Chair: David Lindsay 2:00 PM - 2:15 PM **KAUAI BALLROOM** Session I: AAVP Symposium 2:15-4:00PM Coccidial Biology: Facts and Fiction **KAUAI BALLROOM** 4:15-6:30PM. Session II: Unusual Cases **MAUI BALLROOM** 4:00-6:30 PM Session III: Helminths (I)	**KAUAI BALLROOM** Session IV: 8:00-8:30-Intervet/AAVP Outstanding Graduate Student Talk-Sheila Mitchell Student Papers (I) 8:30 – 9:45 AM	**MAUI BALLROOM** Session V: Arthropods 8:00 - 9:45 AM	**KAUAI BALLROOM** Session: VIII: Heartworms and Helminths (II) 9:00 – 10:00 AM	**MAUI BALLROOM** Session IX: Drug Resistance 8:30 – 10:00 AM	**HAWAII CONVENTION CENTER ROOM 309** Session XI: AAVP/AVMA JOINT SYMPOSIUM Moderator: Ann Donoghue 8:30 AM – 10:00 AM
6:30 – 7:00 PM BREAK	**9:45 – 10:15 AM BREAK**	**9:45 – 10:15 AM BREAK**	**10:00 – 10:30 AM BREAK**		
MOLOKAI BALLROOM BAYER WELCOMING SOCIAL 7:00 – 9:00 PM	**KAUAI BALLROOM** Session IV- Continued Student Papers (I) 10:15 – 11:30 AM	**MAUI BALLROOM** Session V- Continued: Arthropods 10:15 AM – 11:00 AM	**KAUAI BALLROOM** Session VIII- Continued: Heartworms and Helminths (II) 10:30 AM – 12:00	**MAUI BALLROOM** Session IX- Continued: Drug Resistance 10:30 AM – 11:15 AM	
	HILO MEETING ROOM (Second Floor) AAVP Gradute Student Lunch/ Mixer 12:00 – 2:00 PM	**LUNCH (On Your Own)** 12:00 – 2:00 PM	**LUNCH (On Your Own)** 12:00 – 2:00 PM		
	KAUAI BALLROOM Session VI: Protozoa 2:00 – 3:30 PM	**MAUI BALLROOM** Session VII: Giardia and Cryptosporidium 2:00 – 3:00 PM	**KAUAI BALLROOM** Session X: Heartworm and Fleas 2:00 – 3:15 PM		
	3:30 – 4:00 PM BREAK	**3:00 – 3:30 PM BREAK**	**3:15 – 3:30 PM BREAK**		
	KAUAI BALLROOM Session VI-Continued: Protozoa 4:00 – 5:00 PM	**MAUI BALLROOM** Session VII- Continued: Giardia and Cryptosporidium 3:30 – 4:30 PM	**KAUAI BALLROOM** Presidential Address AAVP Business Meeting Awards Session 3:30 – 5:30 PM		
	MOLOKAI BALLROOM MERIAL SPONSORED SOCIAL 6:30 – 9:30 PM		**MOLOKAI BALLROOM FORT DODGE SPONSORED SOCIAL** 6:30 – 9:00 PM		

Program for 2006 AAVP Meeting

SATURDAY JULY 15, 2006

12:00 AM-6:00 PM On site Registration.
Room: Ballroom Foyer (second floor)

8:00 AM-1:00 AM AAVP Executive Board Meeting & Luncheon
Room: Koko Crater (second floor)

2:00 PM-2:15 PM Opening Remarks.
Room: Kauai Ballroom
President: A. Donoghue; Vice President & Program Chair: D. Lindsay

SESSION I
AAVP SYMPOSIUM COCCIDIAL BIOLOGY: FACTS AND FICTION
Room: Kauai Ballroom
Chair: David S. Lindsay

1:30 PM-2:00 PM Presentation Loading

2:15 **1.** Coccidial biology. Do not assume any thing. S.M. Mitchell*, A.M. Zajac, D.S. Lindsay

2:45 **BREAK**

3:00 **2.** Bovine *Eimeria*: What we know and where we stand. A.S. Lucas*, A.M. Zajac, D.S. Lindsay.

3:30 **3.** *Toxoplasma gondii* : complexity of life cycle of this model apicomplexan. J.P. Dubey*

4:00 **BREAK**

Saturday July 15, 2006

SESSION II
UNUSUAL CASES
Room: Kauai Ballroom
Chairs: Louis C. Gasbarre, Alexa Rosypal

4:00-4:15 PM Presentation Loading

4:15 **4.** Encephalitozoonosis in a Pomeranian puppy. K. Snowden*, B. Lewis, K. Logan

4:30 **5.** Characterization of the rate of resolution of pulmonary radiographic lesions following treatment of a cat with paragonimosis. A.S. Peregrine*, S.G. Nykamp, H. Carey.

4:45 **6.** Multiple trematode infections in a young horse. K. Snowden*, M. Buote, B. Porter, T. Norman, D. Schmitz, and T. Craig.

5:00 **7.** An unusual case of nonfatal cytauxzoonosis in a 10-week-old Texas kitten. P.J. Holman, T.M. Craig, E.H. Lee, A. Buchanan, S. Wilson, K.F. Snowden*

5:15 **BREAK**

5:30 **8.** Occurrence of *Taenia saginata* Goeze, 1782 (*Taeniarhynchus saginata*) in a 1.5-year old infant. H.M. Elsheikha*

5:45 **9.** *Mammomonogamus auris* from the middle ears of cats from Saipan. E.G. Tudor, D.G. Armato, D.D. Bowman*

6:00 **10.** Flea circus in horse arena. P.A. Payne*, M.W. Dryden.

6:15 **11.** Flea (*Pulex simulans*) infestation in captive giant anteaters (*Myrmecophaga tridactyla*). A.G. Mutlow, M.W. Dryden*, P.A. Payne

7:00 PM-9:00 PM
BAYER SPONSORED SOCIAL
AAVP Welcoming Social
Room: Molokai Ballroom

Saturday July 15, 2006

SESSION III
HELMINTHS I
Room: Maui Ballroom
Chairs: Wendell Davis, Ann Donoghue

3:30 PM-4:00 PM Presentation Loading

4:00 **12.** Visual diagnosis of *Taenia saginata* cysticercosis during meat inspection: is it unequivocal? S. Abuseir, C. Epe*, M. Kuehne

4:15 **13.** In pursuit of echinococcosis on the Tibet-Qinghai plateau. P.M. Schantz

4:30 **14.** Assessment of *Ascaris suum* egg inactivation and destruction by the vermicomposting worm, *Eisenia foetida*. A. Lucio-Forster*, J.L. Liotta, M. McIntosh, D.D. Bowman.

4:45 **15.** Strongyle egg shedding consistency in horses on farms using selective therapy in Denmark. M.K. Nielsen*, N. Haaning, S.N. Olsen

5:00 **16.** Antigen characterization of paramyosin and protein disulphide isomerase of *Ancylostoma caninum* and its use in immunisation trials. B. Epe*, R. Hüsken, S. Pohl, T.C. Schnieder.

5:15 **BREAK**

5:30 **17.** A comparison of the protein constituents of the major body compartments of the dog roundworm, *Toxocara canis*. T. Sun*, D. Ho, D.D. Bowman

5:45 **18.** The effect of the condensed tannin containing forage, *Sericea lespedeza*, fed as a meal on existing and establishing infection of *Haemonchus contortus* in sheep. L.A. Chafton*, J.E. Miller, J.A. Mosjidis, T.H. Terrill, J.M. Burke

6:00 **19.** Effect of vaccination of goats with H-gal-GP and H11 antigens from intestinal membrane cells of *Haemonchus contortus*. D. D. Olcott*, B.M. Weeks, K. Shakya, W.D. Smith, J.E. Miller

6:15 **20.** Use of the FAMACHA© selective deworming program in hair sheep in Virginia. A. Zajac*, D. Notter, K. Pelzer

7:00 PM-9:00 PM
BAYER SPONSORED SOCIAL
AAVP Welcoming Social
Room: Molokai Ballroom

SUNDAY JULY 16, 2006

8:00 AM-5:00 PM On site Registration.
Room: Ballroom Foyer (second floor)

SESSION IV
STUDENT TALKS
Room: Kauai Ballroom
Chairs: Dan Snyder, Jenni A. Spencer

7:30 AM-8:00 AM Presentation Loading

8:00 Intervet/AAVP Outstanding Graduate Student Talk-Sheila Mitchell

8:30 21. Association between *Babesia bovis* kinete levels in female *Boophilus microplus* and infection rates of larval progeny. J. Howell*, M. Ueti, G. Palmer, D. Knowles

8:45 **22.** Prevalence of *Aelurostrongylus abstrusus* infection in urban cats from the Rio de Janeiro metropolitan area, Brazil. S.L.M. Souza-Dantas*, O.P.M. Basto, N. Labarthr, J. Guerrero

9:00 23. The effect of high hydrostatic pressure processing on *Ascaris suum* eggs. A.C. Rosypal*, D.D. Bowman, D.G. Goodwin, D. Holliman, G.J. Flick, D.S. Lindsay

9:15 **24.** Species-specific detection of equine large strongyle DNA by real-time PCR. M.K. Nielsen*, D.S. Petersen, R. M. Kaplan

9:30 **25.** Development of a multiplex real-time PCR for differentiating and quantifying eggs of *Haemonchus contortus*, *Ostertagia ostertagi*, and *Cooperia* oncophora using fecal eggs. A.F. Harmon*, D.S. Zarlenga, M.B. Hildreth

9:45 **BREAK**

10:15 **26.** Evidence of *Cystoisospora canis* monozoic cyst development in vitro and in mice. S.M. Mitchell*, J.P. Dubey, A.M. Zajac, D.S. Lindsay.

10:30 **27.** Effect of weaning method and transport on coccidia oocyst shedding and species composition in beef calves. A.S. Lucas*, W.S. Swecker, G. Scaglia, D.S. Lindsay, F.C. Elvinger, J.P. Fontenot, A.M. Zajac

10:45 **28.** Biological characterization of *Tritrichomonas foetus* from cats. H. Stockdale*, S. Rodning, D. Givens, J.A. Spencer, R.C. Bird, P. Deinnocentes, C.C. Dykstra, B.L. Blagburn

11:00. **29.** Prevalence of antibodies to *Encephalitozoon cuniculi* in horses from Brazil. D. Goodwin*, S.M. Gennari, D.K. Howe, J.P. Dubey, A.M. Zajac, D.S. Lindsay

11:15 **30.** Serologic detection and molecular characterization of *Angiostrongylus vasorum* in dogs. I. Verzberger*, R.J.F. Markham, J.P. Lewis, J. Sheppard, G.A. Conboy

12:00-2:00 AAVP Grad Student Lunch /Mixer
Room: Hilo Meeting Room

Sunday July 16, 2006

SESSION V
ARTHROPODS
Room: Maui Ballroom
Chairs: Patricia A. Payne, James A. Hawkins

7:30 AM-8:00 AM Presentation Loading

8:00 **31.** An apyrase gene encoding a calcium-dependent, structural homologue of the salivary apyrases from blood feeding arthropods has been identified in *Ostertagia* third and fourth stage larvae. D.S. Zarlenga*, L.C. Gasbarre

8:15 **32.** Organophosphate resistance in hematophagous arthropod parasites of cattle: Acetylcholinesterase mutations in horn flies (*Haematobia irritans*) and southern cattle ticks (*Boophilus microplus*). K.B. Temeyer*, J.H. Pruett, A.C. Chen, P.M. Untalan, K.H. Lohmeyer, L.D. Foil

8:30 **33.** Seasonal questing activity of *Amblyomma americanum* (Acari: Ixodidae) in eastern Kansas. M.W. Dryden*, P.A. Payne, S.N. Dryden, V. Smith, M. Hobson.

8:45 **34.** Adverse reactions to schistosomiasis in outbred albino mice exposed to insecticide contamination. H.M. Elsheikha*, M.A. Wahba

9:00 **35.** Evaluation of imidacloprid (8.8%w/w)+permethrin (44%w/w) and fipronil (9.8% w/w)+(s)-methoprene (8.8%w/w) spot-on topical solutions to repel, prevent attachment and kill adult *Rhipicephalus sanguineus* and *Dermacentor variabilis* on dogs. M.W. Dryden*, P.A. Payne, V. Smith, J. Hostetler

9:15 **36.** Efficacy of imidacloprid (8.8%w/w) plus permethrin (44%w/w) spot-on topical solution against *Amblyomma americanum* infesting dogs using a natural tick exposure model. M.W. Dryden*, P.A. Payne, V. Smith, J. Hostetler

9:30 **37.** Evaluation of imidacloprid (8.8%w/w)+permethrin (44%w/w) and fipronil (9.8% w/w)+(s)-methoprene (8.8%w/w) spot-on topical solutions to repel, prevent attachment and kill adult *Ixodes scapularis* and *Amblyomma americanum* on dogs. M.W. Dryden*, P.A. Payne, V. Smith, J. Hostetler

9:45 **BREAK**

10:15 **38.** BoLA Class IIa microsatellite marker within the DRB3 locus that segregates with the *Amblyomma americanum* resistant phenotype. J.H. Pruett*, P.M. Untalan

10:30 **39.** Characterization of transcripts that play a role in successful tick feeding—identification of potential targets for development of novel control strategies for the Southern cattle tick, *Rhipicephalus* (*Boophilus*) *microplus*. P.M. Untalan*, J.H. Pruett, K.B. Temeyer, R.B. Davey

10:45 **40.** Evaluation of acaricide treatments in the control of *Boophilus microplus* (Canestrini, 1887) in cattle in Pernambuco State. V.L.A Santana*, M.A.G. Faustino, E.V. Korinfsky, L.C. Alves, M.M. Lima

12:00-2:00 LUNCH

Sunday July 16, 2006

SESSION VI
PROTOZOA
Room: Kauai Ballroom
Chairs: Tom Kennedy, Sheila Mitchell

1:30 PM-2:00 PM Presentation Loading

2:00 **41.** Host distribution, morphology, and molecular systematics of a widespread apicomplexan in Hawaiian fishes. M.L. Kent*, C.M. Whipps, T.M. Work

2:30 **42.** Overview of canine Chagas disease in Texas. S. Kjos, T. Craig, K. Snowden*

2:45 **43.** *Babesia gibsoni* infection in American pit bull terriers in Texas. P.J. Holman, E.H. Lee, K.G. Bendele, A. Buchanan, S.Wilson, K.F. Snowden*

3:00 **44.** Comparison between different diagnostic kits used for canine visceral leishmaniasis. L.C. Alves*, A.R. Albuquerque, F.R. Aragao, M. A.G. Faustino, Y.M. Gomes, R.A. Lira, M. Nakasawa, E.L. Servio

3:15 **45.** Comparative genotyping analysis of *Trichomonas gallinae* strains from pigeons and birds of prey. J. Sansano Maestre*, M.T. Gomez-Munoz, M.M. Garijo Toledo

3:30 BREAK

4:00 **46.** Prevalence of eukaryotic microbes and genotyping of *Giardia duodenalis* in lambs under 3 months of age in Spain. C. Navarro Villanueva, M.M. Garijo Toledo, M. V. Dominguez Marquez, M. Jimenez, R. Borras Salvador, M.T. Gomez-Munoz*.

4:15 **47.** Seroprevalence of *Neospora caninum* and *Toxoplasma gondii* in Caribou (*Rangifer tarandus*), Moose (*Alces alces*), Wolf (*Canis lupus*), Fox (Vulpes vulpes), and Coyote (*Canis latrans*) populations in Alaska. E. Stieve, K. Beckmen, S. Patton*

4:30 **48.** Effect of salt water and chemical disinfectants on the sporulation and infectivity of *Toxoplasma gondii* oocysts. A.A. Gajadhar*, L.B. Forbes, L. Measures

4:45 **49.** Investigation of the SnSAG family of surface antigens in *Sarcocystis neurona*. D.K. Howe, R. Gaji, C. Crowdus, A. Marsh, W.J. Saville, D.S. Lindsay, J.P. Dubey, D. Granstrom*

6:30 PM-9:30 PM
MERIAL SPONSORED SOCIAL
Room: Molokai Ballroom

Sunday July 16, 2006

SESSION VII
GIARDIA* AND *CRYPTOSPORIDIUM
Room: Maui Ballroom
Chairs: Aaron Lucas, Doug Hutchens

1:30 PM-2:00 PM Presentation Loading

2:00 **50.** Accurate diagnosis of *Giardia spp.* and proper fecal examination procedures. P.A. Payne*, M.W. Dryden, V. Smith

2:15 **51.** An examination of the shedding of cysts and antigen in the feces of laboratory reared Beagle dogs naturally infected with *Giardia.* D.D. Bowman*, J.L. Liotta, F.C. Davis, K.W. Simpson

2:30 **52.** An up-date on the prevalence of *Giardia* in "symptomatic" dogs and cats throughout the United States as determined by the IDEXX SNAP® *Giardia* Test. D.D. Bowman*, J.M. Scarlett, J.C. Garrett, L. Lorentzen

2:45 **53**. Evaluation of the association between *Giardia* infection and pruritic skin disease in dogs. A.S. Peregrine*, B. Skubic-Vengust, J.A. Hall, R. Reid-Smith, A. Lam

3:00 **BREAK**

3:30 **54.** Risk factors for *Cryptosporidium parvum* infection and disease in Ontario dairy calves. L. Trotz-Williams*, S.W. Martin, K.E. Leslie, T. Duffield, D.V. Nydam, A.S. Peregrine

3:45 **55.** Prevalence of *Cryptosporidium* species and genotypes in a 12-month longitudinal study in dairy calves. M. Santin*, J.M. Trout, R. Faye

4:00 **56.** A comparative study of *Cryptosporidium andersoni* and *Giardia* infections in feedlot cattle of Western Australia and Alberta, Canada. B. Ralston*, R.C. Andrew Thompson, D. Pethick, T. McAllister, M. Olson

4:15 **57.** Evaluation of two simple tests for *Cryptosporidium parvum* in calf feces. L. Trotz-Williams*, S.W. Martin, D. Martin, T. Duffield, K.E. Leslie, D.V. Nydam, F. Jamieson, A.S. Peregrine

6:30 PM-9:30 PM
MERIAL SPONSORED SOCIAL
Room: Molokai Ballroom

MONDAY JULY 17, 2006

8:00 AM-12:00 PM On site Registration.
Room: Ballroom Foyer (second floor)

SESSION VIII
HEARTWORMS AND
OTHER HELMINTHS
Room: Kauai Ballroom
Chairs: Daniel E Snyder, Anne Zajac

8:30 AM-9:00 AM Presentation Loading

9:00 **58.** Equine cyathostomins: secretory and inflammatory status in the ventral colon of naturally infected and maximally treated ponies. T.R. Klei*, R. McConnico, S.R. Chirgwin, J.D. Hubert, S.U. Coleman

9:15 **59.** Controlling gastrointestinal nematode infection in goats using sun-dried forage high in condensed tannins. T.H. Terrill*, G.S. Dykes, S.A. Shaik, J.E. Miller, B. Kouakou, G. Kannan, J.M. Burke, R.M. Kaplan, J.A. Mosjidis

9:30 **60.** Efficacy evaluation of macrocyclic lactone pour-ons under field conditions. K.B. Hart*, D.H. Bliss

9:45 **61.** Efficacy of fenbendazole and milbemycin oxime for treating baboons infected with *Trichuris trichiura.* M.V. Reichard*, R.R. Wolf, D.W. Carey, J.J. Garrett, H.A. Briscoe

10:00 **BREAK**

10:30 **62.** Current status of canine heartworm disease in Korea and its potential public health importance. S. Shin*, S. Kang, J. Kwon

10:45 **63.** Results and realistic implications of the 2004 AHS-Merial heartworm survey. J. Guerrero*, C.T. Nelson, D.S. Carithers

11:00 **64.** Data from the 1992-2002 annual heartworm surveys for Ontario, Canada and effectiveness of preventive medication. O. Slocombe*

11:15 **65.** Clinical and pathologic reactions in dogs of heartworm extract-induced shock. M.A.G. Barbosa*, R.A. Mota, L.C. Alves, F.L.C. Brito, E.R. Albuquerque

11:30 **66.** Effects of ivermectin and doxycycline administered alone, together, or together plus melarsomine against adult *D. immitis* in dogs with induced infections: Clinical observations. M.T. Dzimiznski*, J.W. McCall, R. Roberts, S.D. McCall, L.H. Kramer, C. Genchi, J. Guerrero, A. Mansour, N. Supakorndej, P. Supakorndej

11:45 **67.** Effects of ivermectin and doxycycline administered alone, together, or together plus melarsomine against adult *D. immitis* in dogs with induced infections: Parasitological data. J. W. McCall*, S.D. McCall, L.H. Kramer, N. Supakorndej, A. Mansour, P. Supakorndej, M.T. Dzimianski, R. Roberts, C. Genchi, J. Guerrero

12:00-1:30 **LUNCH**

Monday July 17, 2006

SESSION IX
DRUG RESISTANCE
Room: Maui Ballroom
Chairs: David Goodwin, Byron L. Blagburn

8:00 AM-8:30 AM Presentation Loading

8:30 **68.** Anthelmintic resistance in New Zealand: a canary in a coal mine for US producers? M.J. McArthur*, D.M. Leathwick

9:15 **69.** Macrocyclic lactone-resistant *Oxyuris equi*: Anecdote or emerging problem? C.R. Reinemeyer*, A.A. Marchiondo, J.I. Shugart

9:30 **70.** Continued evaluation of combination anthelmintics treatments in a commercial cattle population where anthelmintic resistant parasites had been observed the previous years. L.L. Smith*

9:45 **71.** Resistance to Gastrointestinal nematodes of cattle: Identification of genomic regions affecting resistance and potential mechanisms. L.C. Gasbarre*, T.S. Sonstegard, R. Li, R. Araujo, C.P. Van Tassell

10:00 **BREAK**

10:30 **72.** Ivermectin and moxidectin cause constitutive and induced overexpression of different P-glycoproteins in resistant *Haemonchus contortus.* A. Roulet, R. Prichard*

10:45 **73.** Application of polymerase chain reaction (PCR) and drug sensitivity testing to compare species composition and anti-coccidial drug resistance in *Eimeria* isolated from vaccine and coccidiostat-utilizing poultry. M.C. Jenkin*, S. Klopp, G. Wilkins, K. Miska

11:00 **74.** Confirmation of ivermectin resistance in a purportedly resistant Canadian isolate of *Parascaris equorum* in foals R.M. Kaplan*, C.R. Reinemeyer, O. Slocombe and M.J. Murray.

12:00 – 2:00 PM LUNCH

Monday July 17, 2006

SESSION X.
HEARTWORMS AND FLEAS
Room: Kauai Ballroom
Chairs: Karen Snowden, Craig Reinemeyer

1:30 PM-2:00 PM Presentation Loading

2:00 **75.** Cytokine responses to experimentally induced *Dirofilaria immitis* infections in cats. J.A. Spencer*, S. Barney, S. Major, A.J. Guarino, E.G. Welles, C.M. Johnson, D.M. Tillson, A.R. Dillon, B.L. Blagburn

2:15 **76.** An examination of the published effects of preventative dosages of macrolide treatment on numbers of circulating microfilariae in dogs with patent heartworm (*Dirofilaria immitis*) infections. D.D. Bowman*, C.J. Torre

2:30 **77.** Pathogenesis of feline heartworm infection. J.A. Spencer*, S. Barney, S. Major, A.J. Guarino, E.G. Welles, C.M. Johnson, D.M. Tillson, A.R. Dillon, B.L. Blagburn

2:45 **78.** Discovery, structure, efficacy, and safety of dinotefuran, a third-generation neonicotinoid. A.H. Ahn*

3:00 **79.** Cat flea susceptibility to imidacloprid: Results of a 5-year monitoring initiative. B.L. Blagburn*, R. Bond, R. Arther, I. Denholm, M. Dryden, J. Hostetler, H. Mehlhorn, N. Mencke, P. Payne, M. Rust, I. Schroeder, M. Vaughn

3:15 PM BREAK

Monday July 17, 2006

3:30 PM-3:45 PM PRESIDENTIAL ADDRESS – Ann Donoghue

3:45 PM – 4:45 AAVP BUSINESS MEETING - Agenda to be provided

4:45 PM - 5:30 PM AAVP AWARDS CEREMONY

Student Travel Awards
Bayer Student Paper Competition Awards
Intervet Graduate Student Award
Distinguished Service Award
Distinguished Veterinary Parasitologist Award

5:30 PM PICTURES

6:30 PM – 9:00 PM
FORT DODGE SPONSORED SOCIAL
Room: Molokai Ballroom

TUESDAY JULY 18, 2006

Session XI
8:30 AM – 10:00 AM
President's Symposium
AAVP/AVMA Joint Meeting
Hawaii Convention Center – Room 309
Moderator: Ann Donoghue

8:00 – 8:30 AM Presentation Loading

8:30 to 9:15 AM 80. David Duffy

9:15 to 10:00 AM 81. Parasites in paradise-impacts of parasitic diseases on Hawaii's native species. Thierry Work*

ABSTRACTS

ABSTRACTS

01

Coccidial biology. Do not assume any thing. S.M. MITCHELL, A.M. ZAJAC, D.S. LINDSAY
DEPARTMENT OF BIOMEDICAL SCIENCES AND PATHOBIOLOGY, VA/MD REGIONAL COLLEGE OF VETERINARY MEDICINE, VIRGINIA TECH, BLACKSBURG, VA.

Coccidial biology is a challenging field of study. We have learned much about the developmental biology of coccidia from in vitro and in vivo studies. The present report will review the current knowledge of in vivo development and how it correlates with in vitro development. Chicken embryos will be compared to cell culture systems. Parasite strains and species will be covered to present the best or most used models. Present knowledge of using in vitro studies to predict drug sensitivities will be examined. Experimental design can adversely or positively affect the observed efficacy of agents against coccidia. We will discuss the reasons for this observation using *Toxoplasma gondii* as a model organism.

02

Bovine *Eimeria*: What we know and where we stand. A. S. LUCAS*, A. M. ZAJAC, D. S. LINDSAY. DEPARTMENT OF BIOMEDICAL SCIENCES AND PATHOBIOLOGY, VA/MD REGIONAL COLLEGE OF VETERINARY MEDICINE, VIRGINIA TECH, BLACKSBURG, VA.

Eimeria are economically important bovine parasites worldwide. This presentation will outline the current understanding of these parasites and highlight the important areas needing further research. There are 13 named species infecting cattle in the US, but only 2, *E. bovis* and *E. zuernii* are considered pathogenic. The pathogenicity of these 2 species may be attributed in part to their sexual development in the large intestine. Others that exclusively colonize the small intestine, most notably *E. alabamensis*, may cause disease in cases of heavy infection. All bovine *Eimeria* are intracellular, host specific parasites that have a direct fecal-oral life cycle. Oocysts are omnipresent in environments where cattle are raised. This continual exposure to infective oocysts results in eventual infection of all animals and endemic stability within herds. Calves less than one year of age are at the highest risk for developing disease, but subclinical infection is prevalent in all animals, including yearlings and mature cows. Subclinical coccidiosis has been associated with decreased production in growing animals due to compromised intestinal function, resulting in lowered feed efficiency. Signs associated with clinical disease result from the sexual stage's destruction of large intestinal epithelium. The primary sign of clinical coccidiosis is diarrhea, but severe cases may include hematochezia, anemia, weight loss, and death. Subsequent growth of animals surviving severe infection is often permanently retarded. Factors associated with intensity of subclinical infection and onset of disease are not clearly understood. Many young animals excrete large numbers of oocysts of pathogenic species and never show clinical signs, while others will develop disease. Environmental and weaning stress is thought to precipitate disease, but attempts to experimentally reproduce stress induced outbreaks have not been successful. The study of bovine *Eimeria* has been ongoing for nearly a century, but extensive research is still needed to understand the complicated dynamic involving host, parasite, and environment.

03

***Toxoplasma gondii* : complexity of life cycle of this model apicomplexan.** J.P. DUBEY* APDL, ANRI, ARS, USDA, BUILDING 1001, BELTSVILLE, MD.

Tachyzoites, bradyzoites (in tissue cysts), and sporozoites (in oocysts) are the three infectious stages of *Toxoplasma gondii.* The prepatent period (time to shedding of oocysts after primary infection) varies with the stage of *T. gondii* ingested by the cat. The prepatent period (pp) after ingesting bradyzoites is short (3-10 days) while it is long (18 days or longer) after ingesting oocysts or tachyzoites, The conversion of bradyzoites to tachyzoites and tachyzoites to bradyzoites is biologically important in the life cycle of *T. gondii* and recent findings of intermediate pp of 11-17 days in cats maybe associated to the ingestion of transitional stages between tachyzoites and bradyzoites. Pathogenicity and infectivity of *T. gondii* varies with the stage of *T. gondii* ingested, the host, and the genetics of the parasite. Oocysts were nonpathogenic to cats, irrespective of the dose and the genetic type and bradyzoites are less infective for the mice but highly infective for the cat. Certain genetic types of *T. gondii* are more pathogenic to mice. Why *T. gondii* infections are more serious clinically in some patients is speculative and there are no data at the present time to indicate that the parasite genotype is a determining factor in higher mammals other than mice.

04

Encephalitozoonosis in a pomeranian puppy. K. SNOWDEN[1]*, B. LEWIS[2] AND K. LOGAN[1]. [1]COLLEGE OF VETERINARY MEDICINE, TEXAS A&M UNIVERSITY, [2]TEXAS VETERINARY MEDICAL DIAGNOSTIC LABORATORY, COLLEGE STATION, TX

Interest has recently focused on the group of intracellular, eukaryotic parasites in the phylum Microspora as "opportunistic" pathogens of zoonotic importance. The microsporidian parasite, *Encephalitozoon cuniculi* has occasionally been reported as a cause of fatal neurologic disease in young dogs. The body of a 4 week old pomeranian puppy was presented for post-mortem evaluation at the Texas Veterinary Medical Diagnostic Laboratory (TVMDL). This puppy was the second of 3 puppies in the litter to die. The household maintained multiple breeding bitches of small breeds. Clinical abnormalities reported by the referring veterinarian included head pressing, depression, shivering and other neurologic signs. Prominent histologic lesions included multifocal subacute nonsuppurative meningoencephalitis and multifocal subacute necrotizing granulomatous nephritis. Microsporidian parasites were visualized frequently in brain, kidney and rarely in liver. Using molecular sequencing of a portion of the ribosomal RNA genes, the identity of the parasites was confirmed as *E. cuniculi* Genotype III. The Type III parasites have also been reported in humans, and demonstrate minor molecular differences from rodent or rabbit isolates. Further study is needed to evaluate the importance of this pathogen in dogs, and the role of dogs in possible zoonotic transmission of the parasite.

05

Characterization of the rate of resolution of pulmonary radiographic lesions following treatment of a cat with paragonimosis. A.S. PEREGRINE[1]*, S.G. NYKAMP[2], H. CAREY[3]. [1]DEPARTMENT OF PATHOBIOLOGY, [2]DEPARTMENT OF CLINICAL STUDIES, UNIVERSITY OF GUELPH, ONTARIO, CANADA, [3]TAVISTOCK VETERINARIANS, TAVISTOCK, ONTARIO, CANADA.

In August 2003 (day 0) a 16-month old female domestic short haired cat presented to a veterinary practice in southern Ontario with a 5-month history of dyspnea, coughing and gagging. On auscultation, lung sounds were mildly increased bilaterally. The animal was eating and drinking normally and was negative

for FeLV and FIV. Radiographic findings revealed six various sized (1-3 cm) nodules distributed multifocally in the lungs. Some of the nodules were uniformly soft tissue in opacity (n=2) while others had a soft tissue rim and were air filled (cavitated) (n=4). Diffusely throughout the lungs there was a bronchial lung pattern. Examination of feces with a standard gravitational flotation technique (SGFT) revealed large numbers of eggs of *Ancylostoma* and *Paragonimus*. The cat was therefore treated with (i) fenbendazole at 28 mg/kg q12hr for 21 days, and (ii) prednisone at 0.6 mg/kg q12hr for 21 days, then 0.6 mg/kg q24hr for 7 days. Within 8 days a dramatic clinical improvement was observed. However, by day 49 the cat began coughing more frequently. Thoracic radiographs at this time revealed 7 pulmonary nodules. However, the lesions were more cavitated than on day 0, and the bronchial lung pattern was slightly improved. Since low numbers of *Paragonimus* eggs were observed with a SGFT the cat was treated again with fenbendazole and prednisone, as before. On day 135, the cat was clinically normal and negative on SGFT. Thoracic radiographs still contained 7 pulmonary nodules and were similar in appearance to those obtained on day 49. On day 221, the cat was clinically normal and negative on SGFT. Radiographs revealed fewer pulmonary nodules (n=5) and a mild bronchial lung pattern that was much less evident. The final follow-up radiograph on day 316 showed almost complete resolution of the pulmonary nodules.

06

Multiple trematode infections in a young horse. K. SNOWDEN*, M. BUOTE, B. PORTER, T. NORMAN, D. SCHMITZ, T. CRAIG. COLLEGE OF VETERINARY MEDICINE, TEXAS A&M UNIVERSITY, COLLEGE STATION, TX

A yearling pinto filly was presented to the Texas A&M University Veterinary Teaching Hospital with a history of chronic weight loss. A CBC revealed marked anemia, moderate hypoproteinemia and low serum iron concentration. A fecal occult blood test was positive, indicating gastrointestinal blood loss. A fecal egg count revealed 140 strongyle-type eggs/gram. Post mortem findings included pale mucous membranes (anemia) throughout the body and a diffuse lack of body fat stores. The liver exhibited severe chronic fibrosing cholangiohepatitis with distention of the intrahepatic and common bile ducts with numerous intraluminal trematodes admixed with reddish black, slightly viscous fluid. The trematodes varied from 2 mm to over 2 cm in length. Histologically, the liver showed periportal and periductular fibrosis. Oblique sections of an adult trematode were observed in a bile duct lumen, and oval yellowish brown parasite eggs (70 x 80 μm) were identified in liver sections. Additionally, the small intestinal lamina propria and submucosa multifocally contained large oval parasite eggs containing miracidia. The adult trematodes were morphologically identified as *Fasciola hepatica*. The intestinal eggs resembled those of a local wildlife schistosome, *Heterobilharzia americana*, and DNA analysis of eggs purified from liver tissue confirmed that diagnosis. To our knowledge, this is the first report of dual trematode infections in a horse in the USA and the first report of *H. americana* in a horse.

07

An unusual case of nonfatal cytauxzoonosis in a 10 week old Texas kitten. P.J. HOLMAN[1], T.M. CRAIG[1], E.H. LEE[1], A. BUCHANAN[2], S. WILSON[2], K.F. SNOWDEN[1*]. [1]DEPARTMENT OF VETERINARY PATHOBIOLOGY, TEXAS A&M UNIVERSITY, COLLEGE STATION, TX, AND [2]GLENWOOD ANIMAL HOSPITAL, TYLER, TX.

Feline cytauxzoonosis is a rapid and often fatal disease caused by the hemoprotozoan parasite *Cytauxzoon felis* vectored by *Dermacentor variabilis* ticks. A 10 week old kitten was diagnosed with cytauxzoonosis upon presenting with fever (41°C), lethargy, inappetence, 15% haematocrit, and a 50% parasitemia characterized by signet ring-like forms on thin blood film examination. The diagnosis was confirmed by

sequence analysis of the small subunit ribosomal RNA (SSU rRNA) gene. Therapy included doxycycline, penicillin, imidocarb diproprionate, and prednisolone, in addition to supportive care. The kitten survived the infection despite concurrent medical problems including atopy and infestation by a *Cuterebra* sp. Specific PCR indicates that the cat remains a subclinical carrier of the parasite more than two years after recovery from clinical disease. Nonfatal cytauxzoonosis is rare and has not been previously reported in a kitten this young. This is also the first report of a nonfatal case in Texas.

08

Occurrence of *Taenia saginata* Goeze, 1782 (*Taeniarhynchus saginata*) in a 1.5-year old infant. HANY M. ELSHEIKHA*. MANSOURA UNIVERSITY, MANSOURA, EGYPT.

A 1.5-year old male infant was presented with vomiting, abdominal pain, weakness, loss of appetite, pruritus ani, fever, and irritability. Complete blood count detected eosinophilia. Examination of two consecutive stool samples revealed the presence of proglottids of *Taenia* spp. Species identification was performed by counting the main bilateral uterine branches in gravid proglottids, which were found to be 14 - 23 and by examining the scolex, which had 4 suckers only without a rostellum or hooks. These and other diagnostic morphometric features were used to distinguish *T. saginata* and *T. solium*. The finding of adult *T. saginata* in infant stool was unexpected because this tapeworm rarely affects infants. Fecal examination was also conducted to household contacts and no one was found to be infected with taeniasis. Wide speculation surrounded the source of infant' exposure to this accidental infection. And how disease transmission occurred to infant born to non-infected mother was not known. The infant might have been acquired a tapeworm infection by accidental ingestion of raw or undercooked, infected (measly) beef contains the larva (*Cysticercus bovis*) of *T. saginata*.. Whether this finding represents a transplacental transmission acquired during pregnancy is unclear and need to be tested. It is possible that early neonatal infection was confused with prenatal infection as prenatal infection with *C. bovis* has been reported in calves. However, possibility of prenatal infection with adult *T. saginata* in humans is highly unlikely. Anthelmintic therapy Niclosamide for Tapeworms infection was prescribed and was suffices to expel a 5 m long whole tapeworm ~18 hr post-treatment. In conclusion, at-risk populations in endemic rural areas where public hygiene is poor, when a child presents with an altered alimentary status; intestinal taeniasis should be considered however young the child might be.

09

***Mammomonogamus auris* from the middle ears of cats from Saipan.** EDGAR G. TUDOR[1], DANIELLE G. ARMATO[2], DWIGHT D. BOWMAN*[2]. [1]PARADISE ANIMAL HOSPITAL, SAIPAN, NORTHERN MARIANAS ISLANDS; AND [2]COLLEGE OF VETERINARY MEDICINE, CORNELL UNIVERSITY, ITHACA, NY.

Mammomonogamus auris is a strongylid nematode that has been reported only twice previously: once in 1934 from Foochow, China (on the coast of the Taiwan Strait) and the second time in 1982 from the Saitama Prefecture of Japan (a commuting suburb of Tokyo). The worms have typically been recovered from the middle ear, *cavum tympanum*, of infected cats. *M. auris* is related to *Syngamus trachea* of birds, and like *S. trachea*, the adults are permanently joined *in copula*. In the China report, 11 of 48 examined cats were found to be infected with more than 24 pairs of worms being observed. In the Japan report, 7 of 124 cats were found to be infected. Cats have been found to have anywhere from 1 to 8 pairs of worms per ear. At the clinic in Saipan, cases are a fairly common occurrence. In the first case, a cat was admitted for a complaint of head shaking, and upon exam, was noted to have minimal erythema of the tympanic membrane with no otitis externa. The cat was sent home with a topical antibiotic/anti-inflammatory otic ointment and reexamined in 10 days. On reexamination the erythema was still present,

but on prolonged examination, the erythematous region was seen to "move". A myringotomy was performed and two 1-cm-long, red, nematodes, and a number of smaller worms, were removed. The cat recovered without incident. All subsequent cases (5-6/year) were asymptomatic and were diagnosed on routine exam. All cases have been treated with a myringotomy and saline flush/negative pressure extraction through a silastic tube. All cases (with 1 exception) were treated post operatively with Tresaderm and Revolution. One patient was given no post operative treatment, and on subsequent examination, this cat was again found to have worms in its middle ear.

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Flea circus in horse arena. P.A. PAYNE*, M.W. DRYDEN. DEPARTMENT OF DIAGNOSTIC MEDICINE/PATHOBIOLOGY, KANSAS STATE UNIVERSITY, MANHATTAN KS.

Ctenocephalides felis, the cat flea, has caused many infestations of domestic animals (dogs, cats, dairy calves) but rarely have been reported in horses. The flea team was called to investigate an infestation on a horse farm. The equine veterinarian refused to return to this facility because of numerous flea bites he had personally endured on his last farm call. It was determined on the first site visit that the three dogs housed in the climate controlled indoor horse area were the primary hosts and the horses were not infested. The 150' x 75' arena provided perfect environmental (temperature and humidity) parameters for larval and pupal developmental stages of the flea. Raccoons and opossums had been observed eating cat food available for the barn cats and were possible sources of *C. felis*. The dogs were severely infested (4086 combed from the Boxer) despite numerous, regular and long term administration of topical treatments and professional premise treatment of the arena. A combination of topical, oral and premise treatments eliminated this infestation in 58 days.

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Flea (*Pulex simulans*) infestation in captive giant anteaters (*Myrmecophaga Tridactyla*). ADRIAN G. MUTLOW[1], MICHAEL W. DRYDEN[2]*, PATRICIA A. PAYNE[2]. [1]ROLLING HILLS WILDLIFE ADVENTURE, 625 NORTH HEDVILLE ROAD, SALINA, KS. [2]DEPARTMENT OF DIAGNOSTIC MEDICINE AND PATHOBIOLOGY, COLLEGE OF VETERINARY MEDICINE, KANSAS STATE UNIVERSITY, MANHATTAN, KS.

A pair of captive adult giant anteaters (*Myrmecophaga tridactyla*) became heavily infested with fleas (*Pulex simulans*). *P. simulans* is commonly found on opossums (*Didelphis virginiana*) and raccoons (*Procyon lotor*) in Kansas, but in this case they were shown to be completing the entire life cycle with the anteaters as the host. A single treatment of topical imidacloprid, coupled with removal and replacement of infested bedding was rapidly effective at controlling the infestation and no adverse effects of the drug were noted. The fleas were also observed feeding on a pair of aardvarks in the same building but these animals were not treated and the infestation of these animals ended following control in the anteaters. There have been no previous reports of flea infestation of giant anteaters or aardvarks in the wild or in captivity.

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Visual diagnosis of *Taenia saginata* cysticercosis during meat inspection: is it unequivocal? S. ABUSEIR[1], C. EPE*[2], M. KUEHNE[1]. [1]CENTRE FOR FOOD SCIENCE. INSTITUTE FOR FOOD QUALITY AND SAFETY. UNIVERSITY OF VETERINARY MEDICINE HANNOVER, GERMANY, [2]CENTRE FOR INFECTIOUS MEDICINE. INSTITUTE FOR PARASITOLOGY. UNIVERSITY OF VETERINARY MEDICINE HANNOVER, GERMANY.

A total of 267 cysts were gathered from March to December 2004 from two main abattoirs in northern Germany. The cysts were classified by the usual organoleptic methods during meat inspection as *Cysticercus bovis*. The reported prevalences of cysticercosis in the abattoirs were 0.48% and 1.08%, respectively. The cysts were examined macroscopically for description of their morphology and constituents and classified as viable or degenerative (dead). The DNA was extracted from these cysts and subjected to polymerase chain reaction (PCR) for evaluation of the detection methods used and to make certain that the cysts did indeed belong to *C. bovis* as indicated at the slaughterhouses. Two sets of primers were used with different sensitivity levels. The first, HDP1, was able to detect 200 fg of *T. saginata* DNA, and the other, HDP2, only 1 pg. No more than 52.4% of the samples tested positive for *C. bovis* in the PCR using both primers, while 20% of the viable cysts and 49.2% of the degenerative cysts tested negative with both primers.

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In pursuit of echinococcosis on the Tibet-Qinghai plateau. PETER M. SCHANTZ, DIVISION ON PARASITIC DISEASES, CCID, CENTERS FOR DISEASE CONTROL AND PREVENTION, ATLANTA, GA.

Since, 1997, an international group of parasitologists has been engaged in investigation of echinococcosis in Tibetan populations in the Tibetan-Qinghai plateau. These investigations have documented the presence of the highest human prevalences of both major forms of echinococcosis (cystic (*E. granulosus*) and alveolar (*E. multilocularis*). Dogs are the common final hosts for both species with high rates of infection (20%-30%) of both species in owned and "stray" dogs within the villages. A third, previously undescribed species of *Echinococcus* was discovered in the Tibetan fox. This species has been designated *E. shiquicus* and its possible public health significance remains unknown. Among 3199 people from Shiqu County, Sichuan Province, screened for echinococcosis by portable ultrasound combined with serology (ELISA and immunoblot with recombinant Em18 and hydatid cyst fluid antigen, respectively) it was determined that both alveolar (AE) and cystic (CE) forms of echinococcosis were co-endemic in this area with some of the highest village prevalence values recorded anywhere in the world. The total prevalence of echinococcosis was 12.9%, of which 6.2% were AE and 6.8% were CE. Infections were significantly higher in women than in men. Cases of both CE and AE with lesions of maximum diameter more than 10cm accounted for 30% of patients. Prevalence of both AE and CE was significantly higher in females than in males and increased significantly with age. Pastoralist herdsmen appeared to have the highest risk of contracting the disease with a total prevalence of 19.0%, of which AE reached 9.5%. Village-based surveys in 5 townships revealed that the village prevalence of human AE varied from 0% to 14.3%, whereas human CE prevalence ranged from 0% to 12.1%. The results disclosed a trend of decreasing AE prevalence from 9.4% to 0.9% in these 5 townships from north to south. In addition, investigation by questionnaire indicated that the infection status of a household was closely related to the number of owned dogs and the frequency of contact with dogs, whether hands were washed before meals, and source of drinking water.

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Assessment of *Ascaris suum* egg inactivation and destruction by the vermicomposting worm, *Eisenia foetida*. A. LUCIO-FORSTER*, J.L. LIOTTA, M. MCINTOSH, D.D. BOWMAN. DEPARTMENT OF MICROBIOLOGY AND IMMUNOLOGY, CORNELL UNIVERSITY, ITHACA, NY.

The use of earthworms to treat organic wastes generated by human and animal populations is a method called vermicomposting. This process has many proven advantages including the ability to render nutrients more readily available to plants, increasing the water retention of soils and decreasing the volume of organic material. However, vermicomposting must take place at temperatures below 35°C in order to maintain worm viability. This has the disadvantage that it does not reach the 55°C temperatures required by law for pathogen inactivation in composting systems. Pathogen reduction in the composting process is in large part time and temperature dependent, so it is difficult to understand how vermicomposting can produce a pathogen free product. Although it seems likely that earthworms may inactivate various bacterial and viral pathogens, there is some concern based on prior literature that they may not successfully inactivate the eggs of helminths present in some farm manures or those that find their way into municipal biosolids. Thus, this work looked at the ability of *Eisenia foetida* worms to kill *Ascaris* eggs in potting soil. Over a six-month period, there was no significant (α= 0.05, p=0.563) decrease in the mean number of eggs recovered from soil containing worms sampled at 1, 5, 11, 13, 14, 18, 28, and 183 days after egg addition. In terms of viability, at 1 week after the eggs and worms were placed together, there was no significant reduction (α= 0.05, p value = 0.272) in the mean egg viability with or without worms being present (91.8% *vs.* 93%, respectively). After a period of 6 months in the presence of *E. foetida* earthworms, the mean viability of the eggs recovered was 77.2%. In this study, *E. foetida* earthworms did not achieve a one-log reduction in *A. suum* numbers or viability.

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Strongyle egg shedding consistency in horses on farms using selective therapy in Denmark. M.K. NIELSEN[1]*, N. HAANING[2], S.N. OLSEN[1]. [1]DEPARTMENT OF LARGE ANIMAL SCIENCES, ROYAL VETERINARY AND AGRICULTURAL UNIVERSITY, 48 DYRLAEGEVEJ, DK-1870 FREDERIKSBERG C., DENMARK. [2]CENTRE FOR BUSINESS AND TRAINING, 90 VRANDERUPVEJ, DK-6000 KOLDING, DENMARK.

Knowledge of the consistency in shedding of strongyle eggs over time in individual horses will enable veterinarians to develop optimal parasite control strategies based on the needs of individual patients. This study evaluated the shedding of strongyle eggs over a three year period in 424 horses on 10 farms when a selective anthelmintic treatment regime was used. Faecal egg counts (FEC) using the McMaster method were performed twice yearly, in the spring and the fall, and horses with FEC greater than or equal to 200 eggs per gram (EPG) of faeces were treated. The results are presented as probabilities of the egg count outcome, when two previous egg counts are known. A horse with no strongyle eggs detected in the two previous faecal examinations had an 82 % probability of a zero count, and a 91 % probability of a count below 200 EPG in the third examination, even though no anthelmintic treatments were administered any time during this period (12 months). A horse with two previous FEC of greater than zero, but less than 200 EPG had an 84 % probability of being below 200 EPG the third time as well. When FEC exceeded 200 EPG on the previous two counts, the probability for a horse exceeding 200 EPG on the third count was 59 %, even though anthelmintic treatments were administered on

both previous occasions. In conclusion, these data demonstrate consistency in faecal egg shedding from one grazing season to another in a majority of horses, regardless of whether horses were treated with anthelmintic or not. These data strongly support the concept of targeted selective anthelmintic therapy as a means to provide effective strongyle control in horses. Targeted selective therapy has the additional benefit of delaying the development of anthelmintic resistance.

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Antigen characterization of paramyosin and protein disulphide isomerase of *Ancylostoma caninum* and its use in immunisation trials. C. EPE, R. HÜSKEN, S. POHL, T. SCHNIEDER. INSTITUTE OF PARASITOLOGY, HANNOVER SCHOOL OF VETERINARY MEDICINE, GERMANY.

A protein disulphide homologue of the canine hookworm *A. caninum* was identified by immunoscreening against affinity-purified dog antisera and in vitro expression of a recombinant antigen of ca. 38kDa earlier. The full length sequence was revealed after multiple 3′- and 5′-RACE of the recombinant homologue identified earlier. The cDNA sequence of the PDI described here contains 1784 bp, the genomic sequence 2847 bp with 4 exons of 431bp, 104 bp, 425 bp and 170 bp, respectively. The corresponding amino acid sequence is 493 aa long with a molecular mass of 54783.1. Identities to other (nematode) PDIs after blast analysis are presented, the *A. caninum* PDI is defined as secreted PDI isoform. In recent experiments paramyosin was identified by screening of cDNA libraries with affinity purified dog antisera and sequenced. The cDNA of the paramyosin gene shows an open reading frame of 2631 bp. This sequence shows high identities with paramyosin genes of *C. elegans* (93%), *D. immitis* (90%) and *B. malayi* (90%) in NCBI blast search. Paramyosin was expressed as GST fusion protein of approximately 100 kDa in a GST-labelled expression vector. Immunisation trials with cDNA as well as with the recombinant protein or combinations and with different adjuvants are supposed to give information about its immunogenic properties and its protective potential which proved to be promising in preliminary trials. These trials are presented, giving a putative protection of >50% in challenge infection. Further use in immunisation studies is discussed.

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A comparison of the protein constituents of the major body compartments of the dog roundworm, *Toxocara canis*. TIFFANY SUN*, DEAN HO, DWIGHT D. BOWMAN. COLLEGE OF VETERINARY MEDICINE, CORNELL UNIVERSITY, ITHACA, NY.

Toxocara canis is an ascaridoid parasite of dogs that can causes intestinal obstruction, anemia, and respiratory disease. The parasite can be transmitted from the bitch to her puppies via transplacental infection; so, puppies are often born infected. The parasite's eggs contaminate the environment and serve as a source of infection in humans and other paratenic hosts, causing larval toxocariasis, a form of visceral larval migrans. In order to better characterize the antigens comprising the complete worm, this work characterized antigens from different compartments of adult male and female worms: body wall (muscle, hypodermis, nerve cords, and cuticle), reproductive tract, esophagus, and intestine. These antigens were compared to the proteins harvested from whole worms. Dissected tissues of *T. canis* adults from several different canine hosts were ground in Tris-HCl buffer in liquid nitrogen with a mortar and pestle followed by a tissue homogenization or simply by homogenization. Proteins were separated by size using gradient SDS-PAGE electrophoresis. Silver stained gels were used to compare the proteins recovered from batches of worms from different dogs. Western blots were performed to determine the immunogenic characteristics of the proteins that were reacted with antibodies made against the whole *T.*

canis worm and against a high and low molecular weight protein portion of *T. canis*. As expected, variations existed between the proteins of different body parts, with slight variations between sexes. The antigens of worms from different hosts were virtually identical. Western blots with the antibodies made to whole worm and the high molecular weight fraction revealed strong immunogenic binding of certain *T. canis* parts, especially the intestinal tissue, but blots with the antibodies made to low molecular weight moieties demonstrated very little immunoreactivity. The work will next examine the sharing of constituents using antibodies prepared against individual body compartments and excretory secretory antigens.

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The effect of the condensed tannin containing forage, *Sericea lespedeza*, fed as a meal on existing and establishing infection of *Haemonchus contortus* in sheep. L.A. Chafton[1*], J.E. Miller[1], J.A. Mosjidis[2], T.H. Terrill[3], J.M. Burke[4]. [1]Department of Pathobiological Sciences, Louisiana State University, Baton Rouge, LA; [2]Department of Agronomy and Soils, Auburn University, Auburn, AL; [3]Department of Animal Science, Fort valley State University, Fort Valley, GA; [4]USDA ARS, Booneville, AR.

Control of *Haemonchus contortus* has traditionally relied on the use of anthelmintics, but due to resistance and consumer pressure to reduce use of chemicals in agricultural products, more natural and acceptable alternative approaches are needed. Grazing sericea lespedeza (SL), a condensed tannin containing forage, and feeding it as hay has reduced FEC in sheep and goats. This study was conducted to determine the effect of a ground meal form of SL on *H. contortus* infection in lambs. Twenty-eight mixed sex lambs with essentially zero FEC were randomly allocated to 4 treatment groups (7 animals each). Two groups received a bolus of 5000 *H. contortus* L3 once and the infection was allowed to mature over five weeks (existing infection). The remaining 2 groups received trickle infections of 500 *H. contortus* L3 3 times a week for 3 weeks (establishing infection). SL meal was fed over a 5 week period to one each of the existing and establishing infection groups while the other 2 groups were fed bermudagrass hay (control). After the 5 week feeding period, all groups were fed bermudagrass hay for an additional 2 weeks. FEC was significantly ($p<0.05$) reduced in the existing infection SL fed group over the feeding period. Similarly, FEC was lower in the establishing infection SL fed group, but the difference was not significant ($p>0.05$). After SL feeding was stopped, FEC increased in both infection groups which indicated an effect on female worm fecundity. At necropsy, there were fewer worms in both SL fed groups, but the differences were not significant ($p>0.05$). This trend of fewer worm numbers suggested that there may have been an effect on reducing infection level also. Results indicate that SL meal reduced FEC of existing infections more so than establishing infections of *H. contortus*.

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Effect of vaccination of goats with H-gal-GP and H11 antigens from intestinal membrane cells of *Haemonchus contortus*. DONYA D. OLCOTT[1]*, BRANDON M. WEEKS[1], KRISHNA SHAKYA[1], W.D. SMITH[2], JAMES E. MILLER[1]. [1]DEPARTMENT OF PATHOBIOLOGICAL SCIENCES, LOUISIANA STATE UNIVERSITY, BATON ROUGE, LOUISIANA, AND [2]MOREDUN RESEARCH INSTITUTE, PENICUIK, MIDLOTHIAN, UK.

Extracts of adult *Haemonchus contortus* were purified and used as a vaccine against the blood feeding parasite in goats as previously studied in sheep. The proteins used were H11 and H-gal-GP, hidden gut antigens from the microvillar membrane of the gut of the worm and combined with Quil A as adjuvant, then administered to a group of goats kept on concrete then artificially infected with *H. contortus*. The control group received Quil A injections and also infected. This study was performed to analyze the

effects of the H11/H-gal-GP vaccine when given to goats as compared to sheep. The trial showed that IgG levels peaked three weeks after the first vaccine and remained high throughout the remaining booster series but began to wane after artificial infection. However, the IgG levels remained significantly higher than in the controls throughout the entire study. Overall mean fecal egg counts (FEC) were significantly higher in the controls and packed cell volume levels were significantly higher in vaccinated goats compared to controls from Week 3 post infection to the end of the study. A booster vaccine given Week 7 pi caused a sharp increase in IgG levels, elimination of worm burdens and decrease in FEC in the vaccinated group. 96% fewer *H. contortus* adults recovered at necropsy in vaccinated group compared to controls and >96% reduction in FEC after booster vaccine given during established infection. This study shows that the H11/H-gal-GP vaccine was sufficient in protecting goats after challenge infections but is shorter lived than when given under the same conditions in sheep. Booster vaccine given when infection levels are rising are effective in eliminating infections, reducing FEC and therefore may be used in place of an anthelmintic to control haemonchosis in goats as in sheep.

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Use of the FAMACHA© selective deworming program in hair sheep in Virginia. A. ZAJAC*[1*], D. NOTTER[2], K. PELZER[1]. [1]VIRGINIA/MARYLAND REGIONAL COLLEGE OF VETERINARY MEDICINE AND [2]DEPARTMENT OF ANIMAL AND POULTRY SCIENCES, VIRGINIA TECH, BLACKSBURG VA.

The FAMACHA© selective deworming program for haemonchosis in small ruminants has been enthusiastically embraced by producers in the southeastern U.S. At the same time, the popularity of hair sheep is growing in this region. This study was undertaken to compare the effects of the FAMACHA© system and a traditional anthelmintic control program on weight gain and anthelmintic use in a group of predominantly hair and hair cross lambs. Fifty-four St. Croix. hair cross and Suffolk ewes and their spring-born lambs were divided into 2 groups and pastured on approximately equivalent pastures naturally contaminated with strongylid nematodes at the Virginia Tech Sheep Center. In May all ewes were dewormed with levamisole. Until weaning in August, ewes were weighed and scored by the FAMACHA© system at 2 week intervals. Blood and fecal samples were also collected. Lambs were also weighed, sampled and FAMACHA© scored beginning in June and continuing 1 month beyond weaning. One group of ewes and lambs received monthly deworming treatments during the study, which has been standard management practice for this flock. The second group was dewormed based on FAMACHA© score (lambs with a score of 3 or lower were dewormed). None of the ewes in the FAMACHA group required treatment. Individual lambs in the FAMACHA© group were given a score of 3 or below 21 times and were dewormed. Evidence of clinical haemonchosis was not observed in either group. Mean FAMACHA© scores and Packed Cell Volumes were significantly lower in the FAMACHA© lambs, but remained in the normal range. Lamb weights were similar in the 2 groups. The FAMACHA© deworming program appears to be well suited to Caribbean hair and hair cross sheep that often need less intense management for parasitism than more susceptible breeds.

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Association between *Babesia bovis* kinete levels in female *Boophilus microplus* and infection rates of larval progeny. J. HOWELL[1]*, M. UETI[1], G. PALMER[1], D. KNOWLES[1,2]. [1]DEPARTMENT OF VETERINARY MICROBIOLOGY AND PATHOLOGY, WASHINGTON STATE UNIVERSITY, PULLMAN, WA, AND [2]ADRU/ARS/USDA, PULLMAN, WA.

The prevalence of babesiosis and the *Boophilus* tick vector that transmits it is a growing concern to the U.S cattle industry. Widespread acaracide resistance and importation of infected cattle increase the likelihood of disease reemergence. Research defining the transmission dynamics is necessary to better understand the risk imposed by entry of infected cattle in association with competent tick vectors. The objective of this study was to determine how kinete infection levels in hemolymph of exposed *Boophilus* females detected by both light microscopy (LM) and nested PCR correlated with infection rates of larval progeny, responsible for subsequent transmission to naïve cattle. Splenectomized Holstein calves were infested with normal *B. microplus* larvae and inoculated intravenously with the T2Bo strain of *B. bovis*. Replete female *B. microplus* were collected daily during ascending parasitemia. Hemolymph was examined by LM on day 7-10 post repletion. Females were scored based on kinete levels as >10 kinetes/hpf, 1-9 kinetes/hpf, or no detectable kinetes. Samples of hemolymph from females without LM detectable kinete infection were tested by nested PCR using primers targeting *msa-1*. Hemolymph from 32 females were positive by PCR (negative LM/positive PCR) and 30 were negative (negative LM/negative PCR). Eggs from each group of females were kept separate and allowed to hatch. Approximately 1 g each of moderately infected (1-9 kinetes/hpf), negative LM/positive PCR, and negative LM/negative PCR larvae or 0.5g of highly infected larvae (>10 kinetes/hpf), were then applied to susceptible splenectomized calves. Larvae were removed daily for the first four days and infection rates were determined by PCR. Although infection may be undetectable by LM in hemolymph, *B. bovis* exposed females can transovarially transmit *B. bovis* to their offspring, which can subsequently transmit the parasite causing clinical disease. These females may represent the majority of vectors transmitting *B. bovis* under natural conditions.

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Prevalence of *Aelurostrongylus abstrusus* infection in urban cats from the Rio de Janeiro metropolitan area, Brazil.
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Aelurostrongylus abstrusus is the most commonly diagnosed lungworm of domestic cats. It is frequently the etiological agent of asymptomatic infections, but also of mild to severe clinical cases. Severe symptoms of respiratory disease are most common in the young, immunosuppressed, weakened or those cats affected by concomitant infections. In the Brazilian clinical routine, diagnosis of this infection is sparse. Generally it is accomplished by necropsy although its L1 can be easily found in infected animals'

stools by the Baermann technique. Aiming to elucidate the prevalence of aelurostrongylosis in cats of Rio de Janeiro metropolitan area, fecal examinations were done in 546 cats using the Baermann technique. The majority of the animals examined were adults (2 to 7 years old) (37.7% - 206/546), females (54.6% - 298/546) and regarding to life style, most of the cats studied were partially kept outdoors (43.8% - 239/546). Twenty cats (3.7% - 20/546) eliminated larvae of *A. abstrusus*. All the samples from cats that eliminated larvae were positive, showing that serial fecal examinations did not increase the sensitivity of the diagnosis. Males and females eliminated larvae in similar proportion and life style of the animals interfered in the prevalence levels, since cats with access to open areas were the ones infected. There was no difference between the prevalence level among animals with an outdoors life style (4.4% - 4/91) or the ones kept partially outdoors (6.7% - 16/239); however there was a difference among them both (6.1% – 20/330) and the indoor cats (0/216). The results of this study demonstrated that access to external areas endanger the animals of being infected suggesting that veterinarians should include this parasite among the diagnostic possibilities for cats who have unspecific respiratory signs, mainly if they have access to open areas of the environment.

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The effect of high hydrostatic pressure processing on *Ascaris suum* eggs. ALEXA C. ROSYPAL[1,2*], DWIGHT D. BOWMAN[3], DAVID G. GOODWIN[1], DANIEL HOLLIMAN[1], GEORGE J. FLICK[1], DAVID S. LINDSAY[1]. [1]VIRGINIA TECH, BLACKSBURG, VA, [2]UNIVERSITY OF NORTH CAROLINA AT CHAPEL HILL, CHAPEL HILL, NC, [3]CORNELL UNIVERSITY, ITHACA, NY.

High hydrostatic pressure processing (HPP) has been shown to be an effective non-thermal means of inactivating non-spore forming bacteria and some protozoa from various food products. Little information is available regarding the effects of HPP on metazoan parasites. Outbreaks of food-borne disease have been associated with importation of food contaminated with fecal material. *Ascaris suum* is used as a surrogate model metazoan parasite for the human roundworm, *Ascaris lumbricoides*, to study the effects of treatments on the inactivation of eggs in sludge. The present study was conducted to determine the effects of HPP on *A. suum* eggs. Unembryonated *A. suum* eggs were collected from sewage and examined prior to HPP treatment. Eggs were subjected to 138 to 552 Mpa for 10 to 60 seconds in a commercial HPP unit. Embryonation was induced after HPP treatments by incubating eggs in 0.01 N sulfuric acid at room temperature. After 21 days, 100 eggs were examined per treatment using a light microscope and the percent of embryonated eggs was determined. Morphological alterations were not observed in the HPP treated eggs. Embryonation was induced in 38-76% eggs that were subjected to 138 and 270 Mpa. No embryonation was observed in eggs exposed to pressures of 241 Mpa or more for 60 seconds or in eggs exposed to 276 Mpa for 10 to 30 seconds. These results indicate that HPP treatment could be used to protect contaminated food items by inactivating *A. suum* eggs and may also have potential in reducing food-borne illness resulting from fecal contamination. This study was financially supported in part by an Animal Health and Disease grant to DSL and GJF.

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Species-specific detection of equine large strongyle DNA by real-time PCR.

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Equine large strongyles (*Strongylus vulgaris, S. edentatus* and *S. equinus*) are important nematode pathogens of horses with high pathogenic potential. Historically, these parasites were the principal parasitic pathogens of horses, but many years of intensive anthelmintic use has greatly decreased the prevalence of these parasites, and reports of disease from these parasites are now rare. However, new national policies in Denmark aimed at reducing the problems of drug resistance in equine small strongyle (cyathostomin) parasites may inadvertently lead to higher levels of large strongyle infections. Presently, the only means of differentiation between strongyle species is a two-week larval culture with subsequent identification of third stage (L3) larvae by a trained technician. But even with a trained technician, sensitivity is poor when *Strongylus* spp. L3 are present in low numbers. The aim of the present study is to overcome this diagnostic problem by developing a real-time Polymerase Chain Reaction (PCR) assay capable of species-specific identification of these three *Strongylus* species. Species-specific primers and TaqMan® probes were designed by alignment of published ribosomal DNA sequences of the second internal transcribed spacer of cyathostomin and *Strongylus* spp. nematodes. Sensitivity and specificity of the primers were tested using traditional PCR. Concentrations of primers and probes were optimized in real-time PCR using primer and probe matrices. The present assay can reliably and specifically detect minute quantities of DNA of each of the three *Strongylus* species whether alone or mixed in the same sample. The current assay can be used for qualitative detection of *Strongylus* spp. eggs in fecal samples, but with further development it has potential for species-specific relative quantification of eggs within a sample.

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Development of a multiplex real-time PCR for differentiating and quantifying eggs of *Haemonchus contortus*, *Ostertagia ostertagi*, and *Cooperia oncophora using fecal eggs*

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Despite the development of PCR assays for identifying cattle nematodes, there remains a need to rapidly quantitate eggs in mixed infections of economically important trichostrongyles. Herein, we describe progress towards the development of a multiplex, real-time quantitative PCR (QPCR) assay for both identifying and quantifying *Haemonchus, Ostertagia* and *Cooperia* eggs. To this end, we modified previously described PCR primers and probes specific for cattle species for use in a multiplex QPCR, then tested them to insure the absence of cross reactivity. Control plasmids were created for each amplified QPCR product and used to assess the linearity and efficiency of amplification, and the potential for PCR inhibition or variation when used on naturally-derived egg DNA. As a trial case, QPCR was performed in conjunction with an optimized DNA extraction protocol using eggs from an animal infected

with *H. contortus*, only. When QPCR was performed on purified plasmid DNAs, linear amplification resulted where the R-square was between 0.98 and 0.99 and the efficiency ranged from 92% to 115%. However, when DNA from *H. contortus* eggs was tested, the R-square remained at 0.98 but the efficiency dropped to 28%. This cause of this reduced efficiency is not known, but could be the results of PCR inhibitors or from target accessibility in a manner that does not necessarily affect the linearity of the amplification curve. Results presented here suggest that multiplex QPCR is possible using fecal eggs, but additional PCR modifications may be necessary to enhance the efficiency and thereby the sensitivity of the reaction.

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Evidence of *Cystoisospora canis* monozoic cyst development in vitro and in mice. SHEILA M. MITCHELL[1]*, J.P. DUBEY[2], ANNE M. ZAJAC[1], DAVID S. LINDSAY[1]. [1]DEPARTMENT OF BIOMEDICAL SCIENCES AND PATHOBIOLOGY, VIRGINIA TECH, BLACKSBURG, VA, AND [2]U.S. DEPARTMENT OF AGRICULTURE, BELTSVILLE, MD.

Development of endogenous stages of *Cystoisospora canis* occurs in the small intestine of canines. Extraintestinal stages occur in the mesenteric lymph nodes (MLN) and other tissues of both definitive and paratenic hosts. The biology of extraintestinal stages is not well studied. Monolayers of African green monkey kidney cell (CV-1) and bovine turbinate (BT) cell were grown on coverslips and infected with 1 X 10^5 excysted *C. canis* sporozoites. On various days, cover slips were removed, fixed and stained for light microscopy. Sporozoites were surrounded by a clear parasitophorous vacuole on days 2 and 10 post infection (PI) in both cell types. Development of a cyst wall surrounding the sporozpoites in BT and CV-1 cells was detected by day 15 and 16 PI, respectively. Division of sporozoites was not observed in either cell type. Sporozoite lengths and widths were similar in both cell types from day 2-17 PI and a slight increase in cyst wall size occurred. These stages resemble monozoic cysts found in extraintestinal tissues in paratenic hosts. Transmission electron microscopy is currently being done to further characterize these stages. In addition, we developed a PCR test to examine the MLN and spleens from interferon-gamma gene knock-out mice infected with *C. canis* oocysts for *C. canis* DNA. We were able to detect *C. canis* DNA in some of the MLN samples from these mice.

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Effect of weaning method and transport on coccidia oocyst shedding and species composition in beef calves. A. S. LUCAS[1]*, W. S. SWECKER[1], G. SCAGLIA[2], D. S. LINDSAY[1], F. C. ELVINGER[1], J. P. FONTENOT[2], A. M. ZAJAC[1]. [1]VA/MD REGIONAL COLLEGE OF VETERINARY MEDICINE AND [2]DEPT. OF ANIMAL AND POULTRY SCIENCES, VIRGINIA TECH, BLACKSBURG, VA.

Weaning is often mentioned as a stressful event that can precipitate clinical coccidiosis in cattle. Recently, several weaning methods have been recommended to reduce stress in calves. The objective of this study was to investigate the effect of weaning method and transport on fecal oocyst count (FOC) and species composition. Beef calves (n=108) (190-240 days of age) were randomly allotted to 3 treatments (day 0): a fenceline weaned group where calves could see their dams (FL), a noseclipped group where nursing was prevented (NC), and a control group left with their dams (Ctrl). On day 7, all steers (n=50) were transported 107 miles to Virginia Tech, and all heifers (n=58) were taken to distant fields on the same farm. Rectal fecal samples were collected from all calves on days 0, 7, and 21. Steers were also sampled on days 14, 35, and 49. A group of steers from each of the treatments previously described were fitted with pedometers and intensively observed on days 8-11. The Modified McMaster's test and Wisconsin sugar flotation test were used for quantification and identification of oocysts. No clinical coccidiosis was seen during the study. The geometric mean FOC for all groups was not significantly

different on days 0, 7, and 21. On days 35 and 49 the Ctrl steer's FOC was lower ($P<0.05$) than the FL and NC, respectively. No differences between steer and heifer FOC were detected. Treatment had no effect on species composition, but slight time effects were detected. Behavioral observation showed that traditionally weaned Ctrl steers exhibited higher levels of stress related behavior ($P<0.05$), although this was not reflected in FOC. Similarly, transport appeared to have no effect on FOC. Results indicate that weaning method and transport may have little effect on the dynamics of *Eimeria* infection in well managed beef herds.

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Biological characterization of *Tritrichomonas foetus* from cats. H. STOCKDALE*, S. RODNING, D. GIVENS, J.A. SPENCER, R. C. BIRD, P. DEINNOCENTES, C. C. DYKSTRA, B. L. BLAGBURN. COLLEGE OF VETERINARY MEDICINE, AUBURN UNIVERSITY

Tritrichomonas foetus is a flagellated protozoan parasite that normally resides in the reproductive tract of cattle. Our laboratory maintains 4 isolates of *Tritrichomonas foetus* obtained from the feces of cats with chronic diarrhea. All cats were treated with a combination of metronidazole, fenbendazole and enrofloxacin. Diarrhea resolved and trichomonads were eliminated from 2 of the 4 cats. Of the 2 remaining cats, diarrhea either did not resolve or recurred. Trichomonads either were not eliminated or recurred after treatment in these cats. Structural examination of these *T. foetus* isolates using differential interference contrast microscopy and scanning and transmission electron microscopy indicated that they were identical to *T. foetus* recovered from cattle. Cloning and sequencing of a PCR product (ITS1 gene) revealed varying similarities to both bovine and feline isolates. Bovine transmission studies using both bovine and feline isolates are in progress. Studies to determine prevalences of *T. foetus* in pet feline populations are also under way.

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Prevalence of antibodies to *Encephalitozoon cuniculi* in horses from Brazil. DAVID GOODWIN[A*], SOLANGE M. GENNARI[B], DANIEL K. HOWE[C], J.P. DUBEY[D], ANNE M. ZAJAC[A], DAVID S. LINDSAY[A*]. [A] DEPARTMENT OF BIOMEDICAL SCIENCES AND PATHOBIOLOGY, VIRGINIA-MARYLAND REGIONAL COLLEGE OF VETERINARY, VIRGINIA TECH, VIRGINIA VA, [B]FACULDADE DE MEDICINA VETERINÁRIA E ZOOTECNIA, UNIVERSIDADE DE SÃO PAULO, SÃO PAULO, [C]DEPARTMENT OF VETERINARY SCIENCE, UNIVERSITY OF KENTUCKY, LEXINGTON, KY, AND [D]UNITED STATES DEPARTMENT OF AGRICULTURE, AGRICULTURAL RESEARCH SERVICE, ANIMAL AND NATURAL RESOURCES INSTITUTE, ANIMAL PARASITIC DISEASES LABORATORY, BELTSVILLE, MD.

Encephalitozoon cuniculi is a microsporidian parasite reported from rabbits, rodents, dogs, and other mammals has recently been implicated as a zoonotic pathogen. Infections in humans have only been seen in severally immunocompromised individuals. The horse human bond has existed for many millennia, so examining *E. cuniculi* prevalence in hoses was logical. *E. cuniculi* has also been previously been implicated in the abortion and still-births in horses. The prevalence of this parasite in horses has yet to be well defined. We examined sera from 559 hoses from Brazil for antibodies to *E. cuniculi.* Indirect immunofluorecent antibody (IFA) test and direct agglutination test (DAT) were used. IFA found 79 (14.1%) positive compared to DAG found 70 (12.5%) positive. DAG compared to IFA as the "gold standard" resulted in a sensitivity of 95.3% and a specificity of 96.9%. Our study suggests horses in Brazil are regularly exposed to *E. cuniculi.* Although not an alarmingly high prevalence it still is a potential source of human exposure.

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Serologic detection and molecular characterization of *Angiostrongylus vasorum* in dogs

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Angiostrongylus vasorum is a nematode parasite infecting the pulmonary artery/right ventricle of wild and domestic canids. Potentially fatal cardiopulmonary disease can occur in infected dogs. In North America, there is an endemic infection focus in Newfoundland, Canada. Canids acquire infection by ingestion of terrestrial snails/slugs intermediate hosts. Diagnosis in dogs occurs by detection of first-stage larvae by Baermann fecal examination. A sandwich-ELISA based on rabbit polyclonal antiserum prepared against ground whole-worm adult antigen was developed for the detection of circulating antigen in infected dogs. Test sensitivity/specificity was 92%/100%. Survey work in Newfoundland dogs with signs of cardiopulmonary disease, comparing Baermann fecal results and the sandwich-ELISA, indicated that fecal testing may miss almost half of the infections. Test to test/day to day variation has complicated the clinical usage of the sandwich-ELISA as a diagnostic tool. In order to standardize the test, increase sensitivity, and develop a dependable source of diagnostic antigen(s), we have attempted to characterize the antigen(s) detected in the sandwich-ELISA test. We developed a cDNA library, after screening with rabbit anti-*A.vasorum* serum; we have identified 8 clones and obtained sequence data. Once identified, the most commonly occurring sequence was chosen; it was inserted into a pGEX vector and transformed using BL21 bacteria. Following the transformation, protein was expressed using IPTG. Protein was analysed by SDS-PAGE. We were able to identify the recombinant protein by Western blot using rabbit *A.vasorum*-antisera. This protein will be purified using glutathione S-transferase (GST) affinity tag. Purified protein will then be introduced into mice for production of monoclonal antibodies and tested in our sandwich ELISA. The success of the research will enable early detection of the parasite and will allow treatment of dogs infected with *A.vasorum* before permanent cardiopulmonary damage occurs, making complete recovery possible.

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An apyrase gene encoding a calcium-dependent, structural homologue of the salivary apyrases from blood feeding arthropods has been identified in *Ostertagia* third and fourth stage larvae

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Apyrases (ATP-diphosphohydrolase) comprise a ubiquitous class of glycosylated enzymes involved in hydrolyzing extracellular nucleoside di- and triphosphates to mononucleotides and orthophosphate in the presence of Mg^{2+} and/or Ca^{2+}. To date, most apyrases can be structurally linked to the yeast GTPase/CD39 which is part of the heat shock70/sugar kinase/actin superfamily. This group of proteins has been associated with motility, adhesion, secretion, regulation of hemostasis, non-synaptic information transfer and platelet formation. A second class of soluble apyrases which counteract the blood-clotting mechanisms of the host has been identified in hematophagous arthropods. Herein, we identified an apyrase gene that structurally conforms to the soluble, calcium-activated apyrases found in arthropods, by immunologically screening an *Ostertagia ostertagi* L4 cDNA expression library with hyperimmune bovine serum. Host antibodies to the *O. ostertagi* apyrase (OoAP) were detected as soon as 14 dpi and increased through 30 dpi. Mouse antibodies generated against the recombinant protein (rOoAP) were used for immunohistochemical and Western blot analyses. Studies demonstrated that OoAP is produced or collects within the muscle layer below the hypodermis, and is predominantly found in L3 and minimally expressed in L4. Also, mouse anti-rOoAP recognized conserved amino acid epitopes in L3 antigen from

Haemonchus, *Cooperia*, and *Oesophagostomum*. With respect to activity, rOoAP was found to be calcium-dependent with a pH maximum between 6.5 and 7.5 where the greatest functional activity was observed on ATP, ADP, UTP and UDP. Little or no activity was observed when either CTP or AMP were used as substrates, respectively. Striking correlations will be raised between the ability of extracellular ATP and orthophosphate to regulate muscle development and movement, and the role this class of apyrases may play in the ontogeny of *O. ostertagi*.

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Organophosphate resistance in hematophagous arthropod parasites of cattle: Acetylcholinesterase mutations in horn flies (*Haematobia irritans*) and southern cattle ticks (*Boophilus microplus*). KEVIN B. TEMEYER[1]*, JOHN H. PRUETT[1], ANDREW C. CHEN[1], PIA M. UNTALAN[1], KIMBERLY H. LOHMEYER[1], LANE D. FOIL[2]. [1]KNIPLING-BUSHLAND U.S. LIVESTOCK INSECTS RESEARCH LABORATORY, USDA-ARS, KERRVILLE, TX, AND [2]DEPARTMENT OF ENTOMOLOGY, LOUISIANA STATE UNIVERSITY AGRICULTURAL CENTER, BATON ROUGE, LA.

Organophosphate resistance in arthropods may result from alteration of an acetylcholinesterase. We have identified acetylcholinesterases of *Haematobia irritans* and *Boophilus microplus (Rhipicephalus microplus)* that contain mutations in organophosphate-resistant strains. Baculovirus expression of recombinant acetylcholinesterases demonstrated that these mutations resulted in production of enzymes with reduced sensitivity to organophosphate inhibition. We have developed a number of diagnostic assays for the mutations using genomic DNA-based PCR, allowing detection and assessment of individuals or populations containing the mutations within a few hours, thereby enabling informed decisions for control options.

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Seasonal questing activity of *Amblyomma americanum* (Acari: Ixodidae) in eastern Kansas. M.W. DRYDEN*, P.A. PAYNE, S.N. DRYDEN, V. SMITH, M. HOBSON. DEPARTMENT OF DIAGNOSTIC MEDICINE/PATHOBIOLOGY, KANSAS STATE UNIVERSITY, MANHATTAN KS.

Determining tick questing patterns will help us to provide veterinary practitioners, entomologists and the public basic knowledge of tick activity, an insight to when to seasonally time acaracide application and a general understanding of the behavior of ticks. This project was designed to determine the seasonal questing patterns of *A. americanum,* the Lone Star Tick, in Eastern Kansas. At two week intervals for 12 consecutive months questing ticks of all life stages were collected using a light colored 1 m x 1.25 m flannel tick "drag". The area sampled was a wooded area (primarily Oak canopy) at the Konza Prairie Biological Station. The "drag" was pulled slowly over the ground along 4 - 50 meter drag lines to capture questing ticks. Collected ticks were placed into vials containing 70% alcohol and transported to the laboratory for species and life stage identification. Dragging was initiated on 30 Mar 04 when one male *A. americanum* was recovered. Tick numbers escalated rapidly so that 2 and 4 weeks later 14 adults and 3 nymphs and then 38 adults and 18 nymphs were collected, respectively. Adult tick collections peaked on 25 May when 37 were collected and the last adults were collected on 07 Jul when 4 were recovered from the drag. Nymphs peaked between 07 Jul and 21 Aug when 52 and 46 ticks were collected, respectively. Larvae were first recovered on 18 Jul and the last collections of larvae were on 30 Oct. Larval numbers peaked on 04 Sept when 5,715 were collected.

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Adverse reactions to schistosomiasis in outbred albino mice exposed to insecticide contamination. H.M. ELSHEIKHA*[1], M.A. WAHB.[2]. [1] DEPARTMENT OF PARASITOLOGY, COLLEGE OF VETERINARY MEDICINE; [2] GASTROENTEROLOGY SURGERY CENTER, COLLEGE OF HUMAN MEDICINE, MANSOURA UNIVERSITY, MANSOURA, EGYPT.

The present study tested the hypothesis that mice infected with the digenetic fluke *Schistosoma mansoni* and exposed to the insecticide larvin, have an increased risk of accelerated liver damage. To investigate this hypothesis, adverse effects resulting from exposure to larvin were compared between *S. mansoni*-infected and non-infected outbred albino mice. The effects of concurrent treatment with larvin on the progress and outcomes of *S. mansoni* infection were assessed via macroscopic and microscopic examination of liver and spleen and evaluation of hematological, biochemical and hepatic enzymatic parameters as well as worm burden. Oral administration of larvin at 20- or 10-mg/Kg BW to mice infected with 200, 100 or 50 *S. mansoni* cercariae induced (i) hepatomegaly and splenomegaly; (ii) prominent lymphocytic aggregation in liver replacing large areas of bridging necrosis; (iii) increased serum level of bilirubin and transaminases (GOT and GPT) enzymes; (iv) decreased serum level of albumin and total proteins; (v) decreased RBCs, lymphocytes, leucocytes count, and hemoglobin content; and (vi) no effect on the number of adult worms of *S. mansoni* or oviposition, compared to mice infected with corresponding numbers of cercariae without larvin treatment or non-infected mice treated with 20 mg/Kg or 10 mg/Kg larvin. All doses used for infection of mice with *S. mansoni* cercariae or treatment with larvin resulted in dose dependent alterations in hepatic functions of the tested mice. Such alterations were more severe in mice treated with the higher dose of larvin and/or infected with the largest numbers of *S. mansoni* cercariae. Additionally, these alterations were severely modified when *S. mansoni* infection and larvin treatment occurred together. These findings support our hypothesis and show that concurrent *S. mansoni* infection with exposure to larvin adversely affect liver and spleen and seriously alter hematological, biochemical, and hepatic enzymatic functions in outbred albino mice.

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Evaluation of imidacloprid (8.8%w/w)+permethrin (44%w/w) and fipronil (9.8% w/w)+(s)-methoprene (8.8%w/w) spot-on topical solutions to repel, prevent attachment and kill adult *Rhipicephalus sanguineus* and *Dermacentor variabilis* on dogs. M. W. DRYDEN[1]*, P. A. PAYNE[1], V. SMITH[1], J. HOSTETLER[2]. [1]DEPARTMENT OF DIAGNOSTIC MEDICINE /PATHOBIOLOGY, COLLEGE OF VETERINARY MEDICINE, KANSAS STATE UNIVERSITY, MANHATTAN, KS. [2]BAYER ANIMAL HEALTH, PO BOX 390, SHAWNEE MISSION, KS.

This study evaluated the effectiveness of imidacloprid + permethrin and a fipronil+(s)methoprene spot-on topical solution to prevent attachment and kill adult *R. sanguineus* and *D. variabilis*. Twelve purpose bred beagles were distributed into 3 groups: 4 dogs treated with imidacloprid 8.8%w/w+permethrin 44%w/w spot-on, 4 dogs treated with fipronil 9.8%w/w+(s)methoprene 8.8%w/w spot-on and 4 dogs as untreated controls. Dogs were exposed to 25 adult *R. sanguineus* and 25 adult *D. variabilis* for 10 minutes on days 3, 7, 14, 21 and 28 post-treatment. Ticks not attaching to the dogs were collected and evaluated for viability. Tick counts on dogs were conducted 3, 24 and 48 hrs after tick exposure. A rapid avoidance or "repellent-like" effect of the imidacloprid+permethrin formulation was evident on days 3 – 21 and 3 - 28 post-treatment, respectively for *R. sanguineus* and *D. variabilis*. There was a significant effect on the viability of non-attaching *R. sanguineus* and *D. variabilis* that were exposed to imidacloprid+permethrin treated dogs. The 3 hour post-exposure tick counts conducted on days 3 – 28, were significantly reduced on dogs treated with imidacloprid+permethrin as compared to the control group. Tick counts on imidacloprid+permethrin treated dogs were significantly reduced for both species on days 14 – 28. Both

imidacloprid+permethrin and fipronil+(s) methoprene treated dogs had significant reductions in geometric mean *R. sanguineus* and *D. variabilis* counts at 24 hours post-exposure on days 3 & 7 post-treatment, and significant reductions in geometric mean tick counts at 48 hours post-exposure at every time point post-treatment. The 48 hour efficacy of the imidacloprid+permethrin formulation against *R. sanguineus* and *D. variabilis* on day 28 post-treatment was 100%, and 92.0% respectively. The 48 hour efficacy of the fipronil+(s) methoprene formulation against *R. sanguineus* and *D. variabilis* on day 28 post-treatment was 86.3% and 83.23%, respectively.

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Efficacy of imidacloprid (8.8%w/w) plus permethrin (44%w/w) spot-on topical solution against *Amblyomma americanum* infesting dogs using a natural tick exposure model. M. W. DRYDEN[1]*, P. A. PAYNE[1], V. SMITH[1], J. HOSTETLER[2]. [1]DEPARTMENT OF DIAGNOSTIC MEDICINE /PATHOBIOLOGY, COLLEGE OF VETERINARY MEDICINE, KANSAS STATE UNIVERSITY, MANHATTAN, KS. [2]BAYER ANIMAL HEALTH, PO BOX 390, SHAWNEE MISSION, KS.

This study represents the first account of a natural field exposure model evaluating a topically applied tick product within North America. The objective of this study was to evaluate the efficacy of an imidacloprid 8.8%w/w + permethrin 44%w/w spot-on topical solution against *Amblyomma americanum* using a natural field exposure model. Sixteen beagles were divided into 2 groups of eight dogs. One group of dogs was treated with the imidacloprid+permethrin topical formulation according to label directions, and the other group served as untreated controls. At -1, 3, 7, 14, 21 and 28 days post-treatment, the dogs were walked for 80 minutes in an *A. americanum* infested habitat at the Konza Prairie Biological Station in Northeastern Kansas, USA. The area proved to be heavily infested with ticks as the pretreatment geometric mean *A. americanum* counts were 34.7 and 34.0 ticks/dog for dogs in the control and treated groups, respectively. Post-exposure tick counts (efficacy evaluations) were conducted on each dog at 3 and 48 hours post exposure. At 3 days post-treatment, the efficacy of the imidacloprid-permethrin formulation was 88.0% within 3 hours of natural tick exposure. Efficacy at 3 hours post-exposure declined slowly as the study progressed, with tick efficacy being 64.3 and 68.3% on days 21 and 28, respectively. Efficacy within 48 hours of natural tick exposure against *A. americanum* was 93.5% on day 3, 94.1% on day 21 and 96.6% on day 28 of the study. This natural exposure model demonstrated "real world" results of a topical acaricide (imidacloprid 8.8% w/w + permethrin 44% w/w spot-on) against *A. americanum,* a tick which has dramatically increased its geographic range in the United States.

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Evaluation of imidacloprid (8.8%w/w)+permethrin (44%w/w) and fipronil (9.8% w/w)+(s)-methoprene (8.8%w/w) spot-on topical solutions to repel, prevent attachment and kill adult *Ixodes scapularis* and *Amblyomma americanum* on dogs. M. W. DRYDEN[1]*, P. A. PAYNE[1], V. SMITH[1], J. HOSTETLER[2]. [1]DEPARTMENT OF DIAGNOSTIC MEDICINE /PATHOBIOLOGY, COLLEGE OF VETERINARY MEDICINE, KANSAS STATE UNIVERSITY, MANHATTAN, KS. [2]BAYER ANIMAL HEALTH, PO BOX 390, SHAWNEE MISSION, KS.

The objective of this study was to evaluate the effectiveness of imidacloprid+permethrin and a fipronil+(s)methoprene spot-on topical solution to prevent attachment and kill adult *I. scapularis* and *A. americanum* on dogs. Twelve purpose bred beagles were distributed into 3 groups: 4 dogs treated with imidacloprid 8.8%w/w+permethrin 44%w/w spot-on, 4 dogs treated with fipronil 9.8%w/w+(s)methoprene 8.8%w/w spot-on and 4 dogs as untreated controls. Dogs were exposed to 25 adult *A. americanum* and 25 adult *I. scapularis* for 10 minutes on days 3, 7, 14, 21 and 28 post-treatment. Ticks not attaching to the dogs were collected and evaluated for viability. Tick counts on dogs were

conducted 3, 24 and 48 hrs after tick exposure. A rapid avoidance or "repellent-like" effect of the imidacloprid+permethrin formulation was evident for *A. americanum* on days 3 and 7 and for *I. scapularis* on days 3, 7, 14 and 21 post-treatment.. There was a significant effect on the viability of non-attaching *A. americanum* that dropped off of imidacloprid-permethrin treated dogs at 3, 7 and 14 days post-treatment. Non-attaching *I. scapularis* were remarkably susceptible to both formulations. On days 7 and 21 the geometric mean *A. americanum* counts, at 24 hours post-exposure on the imidacloprid+permethrin treated dogs, were significantly less than on the fipronil+(s) methoprene treated dogs. Both formulations provided significant reductions in *A. americanum* counts at 24 hrs post exposure at each time point from day 3 – 28 post-treatment. The 24 hr post-exposure efficacy of both formulations against *I. scapularis* was similar on days 3, 7 and 14. Dogs treated with imidacloprid+permethrin had significantly reduced 24 hour geometric mean number of *I. scapularis* on days 21 and 28 post-treatment compared to counts on dogs treated with fipronil+(s)methoprene. The 48 hr post-exposure efficacy of both formulations against *I. scapularis* was similar from 7 – 28 days post-treatment.

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BoLA Class IIa microsatellite marker within the DRB3 locus that segregates with the *Amblyomma americanum* resistant phenotype. JOHN H. PRUETT* AND PIA M. UNTALAN. USDA-ARS KNIPLING-BUSHLAND U.S. LIVESTOCK INSECTS RESEARCH LABORATORY, KERRVILLE, TX.

We have established a genomic DNA database representing a herd of cattle that has been phenotyped for susceptibility to ectoparasites. Herd cows and calves have been phenotyped as high or low carriers of the horn fly, *Haematobia irritans irritans*, and calves have been phenotyped for susceptibility or resistance to the Lone Star tick, *Amblyomma americanum*. Genomic DNA isolated from tick phenotyped herd calves (n=98) was amplified using primers designed for three microsatellite (msat) loci on bovine autosome 23 (BTA23) within the bovine major histocompatibility complex (BoLA) and one msat locus that is located on the same chromosome, but not within the BoLA. Specifically, the msat loci DRB3 (Class IIa cluster), MB025 (within bovine DRB1, Class IIa), BM1815 (within DIB, Class IIb cluster), and RM185 (located outside of BoLA) were analyzed using an ABI3100 Genetic Analyzer with the GeneMapper v3.5 software package. Allele frequencies were calculated by direct counting, and association between a particular allele and the phenotype of host susceptibility or resistance was assessed using the Fisher Exact Test. No significant associations were identified at the MB025, BM1815, or RM185 loci. However, allele 174 of the DRB3 locus was significantly associated with the tick resistant phenotype (P=0.003). The DRB3 gene is the most polymorphic within the class II cluster, particularly within exon 2 where 103 alleles have been found (Aida et al., 1995; Davies et al., 1997). The product of this gene is involved in antigen peptide binding and presentation to $CD4^+$ helper cells. We are currently using a PCR-RFLP approach (Ellegren et al., 1993) to identify whether there are variable DRB3-exon 2 sequences between tick susceptible/resistant individuals within the herd.

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Characterization of transcripts that play a role in successful tick feeding—identification of potential targets for development of novel control strategies for the Southern cattle tick, *Rhipicephalus* (*Boophilus*) *microplus*. PIA M. UNTALAN*[1], JOHN H. PRUETT[1], KEVIN B. TEMEYER[1], RONALD B. DAVEY[2]. [1]USDA-ARS KNIPLING-BUSHLAND U.S. LIVESTOCK INSECTS RESEARCH LABORATORY, KERRVILLE, TX AND [2] USDA-ARS, CATTLE FEVER TICK RESEARCH LAB, MOORE FIELD, TX.

The Southern cattle tick, *Rhipicephalus* (*Boophilus*) *microplus*, is an arthropod vector of *Babesia bovis* and *B. bigemina* and *Anaplasma marginale*, the causative agents of bovine babesiosis and anaplasmosis, respectively. Acaricides are currently the primary means of tick control, but the development of tick resistance to these acaricides necessitates the development of alternative control strategies. The identification of molecules that are essential for tick feeding is one approach to the identification of targets for alternative control. Independent cDNA libraries were constructed from salivary gland tissues of adult *R.* (*Boophilus*) *microplus* females that had fed on a naïve bovine host for 1-2 days or 4-5 days in an effort to identify transcripts expressed during at least one of two different feeding stages. Transcripts identified included glycine-rich proteins that have been implicated as critical for formation of the cement cone during tick attachment, metalloproteases, various peptidases, and putative serine proteinase inhibitors. Four transcripts encoding Kunitz-type proteins and five transcripts encoding metalloprotease-like sequences were further characterized, and appeared to be differentially expressed in females versus males and throughout development. Three of the Kunitz-type proteins have been overexpressed in *E. coli* for monoclonal antibody production, while all four of the Kunitz-type transcripts have been expressed in an insect cell system for characterization with respect to the host blood coagulation system.

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Evaluation of ascarides treatments in the control of the *Boophilus microplus* (Canestrini, 1887) in milk bovines kept in the fields in the Pernambuco state. V. L. A. SANTANA[1], M. A. G. FAUSTINO, E. V. KORINFSKY, L. C. ALVES, M. M. LIMA. [1] Ciência Veterinária, UFRPE .

The control of the tick *Boophilus microplus* includes the use systemize of acaricides products on the bovine and, this use has had several consequences for the animal, the farmer and, mainly, the proper tick. It was aimed in this work to evaluate different projects of acaricides treatments for control of the *B. microplus* in liverstock kept in field at Pernambuco state. The first phase of the experiment happened in the collection directly of adult tick of *Boophilus microplus* of the hosts for obtain larvae, followed for three infestations, approximately 2500 larvae, with interval of seven days. In the day the 0 animals had been separate in groups, each one composition four to eight animals, submitted to the same sanitary and alimentary handling, selected for convenience for formation of the experimental groups, as product: Abamectin; Ivermectin; Ivermectin LA; Amitraz; Amitraz + Ivermectin; +Ivermectin Amitraz LA; Amitraz + Abamectin; Association (Cipermetrin + Clorpirifós + Citronela); Association + Ivermectin; Association + Ivermectin LA; Association + Abamectin; Control. In day zero, high infestation for all forms of the tick was evidenced, having itself the predominance of adults. It was verified that, with exception of the groups treated with association and the group controlled had a reduction in the counting of day zero for day +7. The general average of effectiveness of the treat groups demonstrated to reduced indices of effectiveness varying to 31,44%, (for the group ivermectin LA) to 79.06% (for the group amitraz+ivermectin LA). However, to if analyzing the effectiveness per day post-treat, it was verified that these index had gotten a bigger variation (0% - 96.63%), reaching considerable values of effectiveness. Such indices are translating the reduction of the number of tick in the animals, however the effectiveness of the treatments can more good be evaluated, analyzing each animals. Individual study of the animals disclosed difference in the behavior of the infestations for the tick, notifying itself that some animals if had presented with tick more than others of the same group of treatment, or same little infested, demonstrating to the resistance of the host to the attack of the tick, consisting in the best method of control not chemistry of the *Boophilus microplus*.

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Host distribution, morphology, and molecular systematics of a wide-spread apicomplexan in Hawaiian fishes. MICHAEL L. KENT[1], CHRISTOPHER M. WHIPPS[1], THIERRY M. WORK[2]. 1. CENTER FOR FISH DISEASE RESEARCH, COLLEGE, DEPARTMENT OF BIOMEDICAL SCIENCES, COLLEGE OF VETERINARY MEDICINE, OREGON STATE UNIVERSITY, CORVALLIS, OR 97331, 2. UNITED STATES GEOLOGICAL SURVEY, NATIONAL WILDLIFE HEALTH CENTER HONOLULU FIELD STATION, HONOLULU, HI.

We recently described an unusual apicomplexan from the kidney, spleen and liver of ta'ape (*Lutjanus kasmira) in Oahu.* The infection is very common, with surveys showing a prevalence of often over 50%. This fish, also called blue-line snapper, was introduced from French Polynesia to Oahu about 50 years ago, and has since become established in all the Hawaiian Islands. As the fish is an exotic species to Hawaii, there was concern that this parasite may have been introduced with its host and might infect naive fishes in these islands. Because oocysts are not seen, we used both molecular and histological surveys of fishes from Oahu to determine the host range of this parasite. Examination of 5 species of goatfish showed infection prevalence of high, with > 85% in Pfluger's goatfish (*Mulloidichthys pflugeri*) by histology. Apicomplexan SSU rDNA sequence obtained from the kidney and spleen of yellowfin goatfish (*Mulloidichthys flavolineatus*) were identical to those obtained from ta'ape, indicating that they were indeed the same parasite. The same sequence also was obtained from the spleen and kidney of opakakapa (*Pristipomoides filamentosus*) and from the liver of an akule (*Selar crumenophthalmus*). The livers contained sporulated oocysts consistent with *Goussia* spp., suggesting that the parasite might complete its development in this fish. However, we cannot exclude the possibility that the liver had mixed infections with the cryptic ta'ape parasite and oocysts of another *Goussia* sp. Comparison of the SSU rDNA sequence of the ta'ape parasite with those of other apicomplexans also indicated that it was most closely related to *Goussia* spp. It is yet to be determined whether this parasite with broad host specificity is endemic to Hawaii, or was introduced with ta'ape.

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Overview of Canine Chagas Disease in Texas. S. KJOS[1], T. CRAIG[2], K. SNOWDEN[2*]. [1]DEPARTMENT OF ENTOMOLOGY, [2]DEPARTMENT OF VETERINARY PATHOBIOLOGY, TEXAS A&M UNIVERSITY, COLLEGE STATION, TX.

It is well recognized that *Trypanosoma cruzi* is endemic in the United States as evidenced by infections reported in humans, domestic dogs, and a wide range of mammalian wildlife species. Although the disease has been reported in domestic dogs from at least five states, the extent of the disease in this host across the country is not well characterized. Canine Chagas disease usually presents as an acute, often fatal myocarditis in puppies or as a chronic progressive dilitative cardiomyopathy in adult dogs. Several recent canine cases at the Texas A&M University teaching hospital have highlighted the wide range of clinical presentations and demonstrated diagnostic difficulties in confirming parasite infections. Multiple sources including serologic data, case records, and histopathologic cases were compiled in an effort to understand the magnitude of the disease among domestic dogs in Texas. Approximately 500 cases have been documented since 1994, spread across most regions of the state. The distribution of canine cases in Texas closely matches the distribution of the seven insect vector species of *Triatoma* reported from the state.

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***Babesia gibsoni* infection in American pit bull terriers in Texas.** P.J. HOLMAN[1], E.H. LEE[1], K.G. BENDELE[1], A. BUCHANAN[2], S. WILSON[2], K.F. SNOWDEN[1*]. [1]DEPARTMENT OF VETERINARY PATHOBIOLOGY, TEXAS A&M UNIVERSITY, COLLEGE STATION, TX, AND [2]GLENWOOD ANIMAL HOSPITAL, TYLER, TX.

Babesia gibsoni is a hemoprotozoan parasite that causes babesiosis in domestic dogs worldwide. The parasite is reported to be more prevalent in American pit bull terrier (APBT) or American Staffordshire terrier breeds in a number of regions of the U.S. In cases reported in other breeds, often there is documented involvement of dogs of these breeds. Despite the popularity of American pit bull terriers in Texas, to date there are no published cases of canine babesiosis in this state. This present study commenced with two 4-week-old American pit bull terrier puppies in Smith County, Texas that presented with anemia, icterus, and fever. Numerous intraerythrocytic *Babesia gibsoni* parasites were found on thin blood film examination. Although the dam appeared clinically normal (PCV 45%) and no *B. gibsoni* parasites were detectable in a blood smear, diagnostic PCR and parasite *in vitro* culture revealed *B. gibsoni* infection. Eight infected dogs, all APBT, were found during a subsequent screen (thin blood film examination, parasite culture, and *B. gibsoni*-specific PCR) of the 34 cohort dogs (including 26 APBT, 6 American bulldogs, 2 Great Danes) resident at the same kennel and 4 APBT dogs from the same kennel but housed in Louisiana at the time of testing. Screening of 3 additional American pit bull terrier kennels in East Texas found 12 of 26 animals PCR positive for *B. gibsoni.* No infected dogs were detected among more than 150 dogs of other breeds that were also tested from the same geographic region.

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Comparison between different diagnostic kits used for canine visceral leishmaniasis. ALVES, L.C[1]*, ALBUQUERQUE, A.R[1], ARAGÃO, F.R [1], FAUSTINO, M.A.G[1], GOMES, Y.M[2], LIRA, R.A[2], NAKASAWA, M[2], L SÉRVIO, E.[3] [1]UNIVERSIDADE FEDERAL RURAL DE PERNAMBUCO , BRAZIL, [2] CENTRO DE PESQUISAS AGGEU MAGALHÃES/CPQAM, RECIFE-PE, BRAZIL, [3] BIOGENE, BRAZIL.

The definitive diagnosis of canine visceral leishmaniasis (CVL) is obtained through the demonstration of the parasite in bone marrow, spleen, liver and lymph nodes smears, but this diagnostic method is invasive, limited and frequently inconclusive. These disadvantages render serologic methods to be essential to CVL diagnosis. The aim of this work was to compare ELISA systems with different antigens for CVL diagnoses. Serum samples of 142 dogs of varied breeds and ages, of both genders, from Metropolitan region of Recife, State of Pernambuco, Brazil were analized. Two ELISA systems were used in the present work : EIE-Leishmaniose-Visceral-Canina–Bio-Manguinhos, batch 053El0027 which uses soluble promastigotes forms of *Leishmania major-like* and by the Kit of canine calazar ELISA S7®[7], batch 02/05 which uses recombinant peptide of *Leishmania chagasi.* The results obtained in EIE-Leishmaniose-Visceral-Canina–Bio-Manguinhos test with the parasitological criteria revealed sensibility of 65.79% and specificity of 85.58% (CI 95%). The positive and negative predictive values were 62.50% and 87.25% respectively. The analyses performed using the canine calazar diagnostic ELISA S7®kit showed sensibility of 47.37% and specificity of 89.42% (CI 95%). At the parallel association of both tests, sensibility of 81. 99%, specificity of 76.53%, with positive predictive value of 56.07% and negative predictive value of 92.08 %was observed. The results for the series association of both tests, showed a sensibility of 31.16%, specificity of 98.47%, with positive predictive value of 88.19% and negative predictive value of 79.65%.The evaluation in series presented an increase on sensibility and the assessment of the association

in parallel showed an increase in specificity, allowing safer results due to the reduction on false negative and false positive cases.

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Comparative genotyping analysis of *Trichomonas gallinae* strains from pigeons and birds of prey. J. SANSANO MAESTRE*, MT. GÓMEZ-MUÑOZ, MM GARIJO TOLEDO. DEPARTAMENTO DE ATENCIÓN SANITARIA, SALUD PÚBLICA Y SANIDAD ANIMAL. FACULTAD DE CIENCIAS EXPERIMENTALES Y DE LA SALUD. CEU UNIVERSIDAD CARDENAL HERRERA, MONCADA. VALENCIA. SPAIN.

Trichomonas gallinae is the causative agent of trichomoniasis, a disease that affects mainly the esophagus and oropharynx of different species of birds. The parasite produces a granulomatous lesion in the upper digestive tract that makes difficult or impossible the ingestion of food, causing animal death. Pigeons and doves (Columbiformes) are the main hosts and the infection is usually asymptomatic. Raptors which feed on pigeons are very sensitive to contract the clinical form of the disease and they generally die. Trichomoniasis is an important disease of birds of prey in Spain due to habitat destruction that forces raptors to feed on pigeons instead of more usual preys. During a period of one year, we have studied the presence of the flagellated *T. gallinae* in pigeons and birds of prey in the Valencian Community. The presence of *T. gallinae* was evaluated in 457 pigeons and 50 birds of prey coming from different areas of the Valencian Community. Prevalence of the flagellated was 36% (n=166) in pigeons and 10% (n=5) in birds of prey. The techniques employed for diagnosis were direct microscopic observation of crop smears and culture at 37 ºC in a commercial modified Diamond´s medium, which was observed every 24 hours for three consecutive days. DNA was extracted from axenic cultures having > 10^6 cells/ml using a commercial DNA extraction kit (DNeasy® tissue kit, QIAGEN). PCR were performed using TFR1 and TFR2 primers that amplify the 5.8S rRNA gene and the flanking internal transcribed spacer regions, ITS1 and ITS2. Restriction fragment length polymorphism analysis of strains from pigeons and raptors was made using different restriction endonucleases. Results show that there are no differences among strains obtained from pigeons and raptors indicating a high homology in this species.

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Prevalence of eukaryotic microbes and genotyping of *Giardia duodenalis* in lambs under 3 months of age in Spain. C NAVARRO[1] VILLANUEVA; MM GARIJO TOLEDO[1]; MV DOMÍNGUEZ MÁRQUEZ[1,2]; M JIMENEZ [2] R BORRÁS SALVADOR[2], MT GÓMEZ-MUÑOZ[1]*. [1]DEPARTAMENTO DE ATENCIÓN SANITARIA, SALUD PÚBLICA Y SANIDAD ANIMAL. FACULTAD DE CIENCIAS EXPERIMENTALES Y DE LA SALUD. CEU UNIVERSIDAD CARDENAL HERRERA. 46113 MONCADA. VALENCIA, SPAIN. [2]DEPARTAMENTO DE MICROBIOLOGÍA, FACULTAD DE MEDICINA Y ODONTOLOGÍA, HOSPITAL CLÍNICO UNIVERSITARIO DE VALENCIA, AV. BLASCO IBÁÑEZ, VALENCIA, SPAIN.

Giardia duodenalis is a well known flagellate that affects a wide range of host. Lately, a big number of studies have been carried out to elucidate its zoonotic role in domestic animals, although most of them analyze calves and only a few were made in sheep. The reported studies include only *Giardia*, not taking into account other microorganism with zoonotic potential to men. In this study, we have analyzed the role of lambs as carriers of eukaryotic parasites that can also infect man. Two hundred and forty five samples from lambs under 3 months of age were analyzed for the presence of cyst of protist and protozoa in their faeces. The study includes 11 farms and the sampling was made randomly. The number of samples from each farm was proportional to the number of animals per farm to diminish the variability between farms. Faeces were concentrated by filtration, flotation and centrifugation, and then analyzed under a light

microscopy. An IF assay for *Giardia* and *Cryptosporidium* was used (Mer*IF*luor, Meridian Diagnostic). DNA extraction was made using a commercial method (DNeasy® tissue kit, QIAGEN). Genotyping of *Giardia* was carried out by PCR using G7 and G759 primers for B-giardin and posterior sequencing. *Eimeria* and *Blastocystis* were present in every farm (71.4% and 25.7% global prevalence, respectively). *Giardia* and *Cryptosporidum* were present in all farms except one. *Giardia* was shed by 34.7% of the animals (0-70% depending on the farm) and *Cryptosporidium* by 16.7% of the animals (0-28.6%). Other organisms found in faeces using microscopy were compatible with *Entamoeba coli*, *Endolimax nana* and *Entamoeba hartmanni*. When B-giardin sequences of *Giardia* were analyzed, genotype E was the only one present in the analyzed samples. Additional genotyping of other eukaryotic microbes, such as *Blastocystis*, should be made to determine the zoonotic potential of these organisms.

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Seroprevalence of *Neospora caninum* and *Toxoplasma gondii* in Caribou (*Rangifer tarandus*), Moose (*Alces alces*), Wolf (*Canis lupus*), Fox (Vulpes vulpes), and Coyote (*Canis latrans*) populations in Alaska. ERICA STIEVE [1], KIMBERLEE BECKMEN[2], SHARON PATTON[1]*. [1]UNIVERSITY OF TENNESSEE COLLEGE OF VETERINARY MEDICINE, KNOXVILLE, TN AND [2]ALASKA DEPARTMENT OF FISH AND GAME, FAIRBANKS, AL.

Blood collected during the 1998-2004 field seasons from caribou (n=205), moose (n=201), wolf (n=200), fox (n=8), and coyote (n=12) in specific areas of Alaska (USA) was tested for antibodies to *Toxoplasma gondii* with a modified agglutination test (MAT), and for antibodies against *Neospora caninum* with an indirect fluorescent antibody test (IFA). Antibody seroprevalence was 10.7% *N. caninum* and 0.5% *T. gondii* in caribou, 0.5% and 0% in moose, 6.0% and 7.5% for wolf. The 8 fox were negative for both parasites. Perhaps the wolf definitive host is responsible for *N. caninum* transmission to cervids in this region. The role of the fox is unclear. The source of the *T. gondii* infection is unknown, but Dubey et al. *(J. Parasitol.,* 87(2), 2001, p. 439–442) speculated that *Lynx canadensis* could be a possible source. Geographical distribution of species and environment differences between areas in Alaska may alter the spread of these parasites.

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Effect of salt water and chemical disinfectants on the sporulation and infectivity of *Toxoplasma gondii* oocysts. ALVIN A. GAJADHAR[1*], LORRY B. FORBES[1], LENA MEASURES[2]. [1]CENTRE FOR FOOD-BORNE & ANIMAL PARASITOLOGY, CANADIAN FOOD INSPECTION AGENCY, SASKATOON, CANADA, AND [2]FISHERIES AND OCEAN CANADA, LMI, MONT-JOLI, QC, CANADA.

Oocysts of *Toxoplasma gondii* are shed by feline definitive hosts, and survive adverse environmental conditions before and after sporulation. Water contaminated with oocysts has been incriminated in fatal toxoplasmosis of marine and terrestrial mammals and in large outbreaks of human infections. Food animals and wildlife that ingest oocysts in contaminated food and water serve as intermediate hosts and a source of toxoplasmosis for humans. Furthermore, recent episodes of foodborne outbreaks of human cyclosporosis and cryptosporidiosis have raised concerns regarding oocysts contaminating beverages, fruit and vegetables. Strategies for the control of toxoplasmosis involving oocysts include disinfection and prevention of sporulation. The objective of this study was to evaluate the ability of salt water and some commonly used disinfectants in inhibiting the sporulation and infectivity of *T. gondii* oocysts. Tissue cysts from a naturally infected pig were used to infect cats for the generation of oocysts. Freshly passed oocysts were purified and treated with various concentrations of salt water, ethanol, hydrogen peroxide, iodine, Proviodine® and Peroxiguard®. Suspensions of oocysts in petri dishes were gently

agitated at room temperature and examined by microscopy on days 0, 4, 8, 10, 24, 40, 80, and aliquots of 1000 oocysts treated for 8-10 days were tested for infectivity by mouse bioassay. Results indicate that oocysts can sporulate and remain intact in saltwater and disinfectants after several weeks of treatment. The mouse bioassay study confirmed the infectivity of these treated oocysts. This study demonstrates that chemical disinfectants are unreliable for controlling oocysts. Non chemical methods, such as heat and UV treatment should be used in strategies for the control of *T. gondii* oocysts.

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Investigation of the SnSAG Family of Surface Antigens in *Sarcocystis neurona*. D.K. HOWE[1], R. GAJI[1], C. CROWDUS[1], A. MARSH[2], W.J. SAVILLE[2], D.S. LINDSAY[3], J.P. DUBEY[4], D. GRANSTROM[4] *. [1]DEPARTMENT OF VETERINARY SCIENCE, UNIVERSITY OF KENTUCKY, LEXINGTON, KY, [2]DEPARTMENT OF VETERINARY PREVENTIVE MEDICINE, THE OHIO STATE UNIVERSITY, COLUMBUS OH, [3]DEPARTMENT OF BIOMEDICAL SCIENCES AND PATHOBIOLOGY, VIRGINIA-MARYLAND REGIONAL COLLEGE OF VETERINARY MEDICINE, VIRIGNIA TECH, BLACKSBURG, VA, [4]USDA, ARS, BARC, BELTSVILLE, MD.

Sarcocystis neurona possesses a gene family of abundant and immunogenic surface antigens that have been designated SnSAGs. Our research has demonstrated that variation exists in the array of SnSAGs expressed by different isolates of *S. neurona*, which has considerable implications for the development of accurate serologic tests and prospective vaccines for equine protozoal myeloencephalitis (EPM). Most notably, we have found that some parasite isolates do not express SnSAG1, the major surface antigen that was identified in strains UCD-1 and SN3. Of nine EPM isolates of *S. neurona* tested thus far, four were found to be SnSAG1-minus. Additionally, SnSAG1was not detected in a strain isolated from a diseased otter. The SnSAG1-minus isolates were found to possess SnSAG2, SnSAG3, and SnSAG4, thereby confirming that these are *bona fide* strains of *S. neurona*. Since it was possible that other SnSAGs might be present in some strains of *S. neurona*, a search was conducted on a gene sequence database generated from strain SN4, an EPM isolate that lacks SnSAG1. From this analysis, two additional SnSAG genes were identified and designated SnSAG5 and SnSAG6. Currently, we are producing antibodies against recombinant SnSAG5 and SnSAG6, which will allow us to examine the conservation of these new SnSAGs in different *S. neurona* isolates. The data collected thus far demonstrate that isolates of *S. neurona* can differ in their assortment of antigens displayed on the merozoite surface. These results further indicate that there is heterogeneity in the genetic composition of different *S. neurona* isolates, which suggests that phenotypic diversity in traits such as virulence or host preference can exist in these parasite strains.

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Accurate diagnosis of *Giardia spp* and proper fecal examination procedures. P. A. PAYNE*, M. W. DRYDEN, V. SMITH. DEPARTMENT OF DIAGNOSTIC MEDICINE PATHOBIOLOGY, COLLEGE OF VETERINARY MEDICINE, KANSAS STATE UNIVERSITY, MANHATTAN, KS.

A series of investigations evaluated the ability of different testing methods; a swing-head centrifugation technique using two flotation solutions (1.18–specific gravity zinc sulfate and 1.27–specific gravity Sheather's sugar solution), a passive commercial flotation technique, and the SNAP Giardia Test Kit from IDEXX Laboratories, to identify Giardia-positive dogs and recover the eggs of other intestinal parasites. In the trials evaluating repeated fecal examinations from research beagles, a single $ZnSO_4$ fecal flotation with centrifugation was positive 37.5% to 87.5% of the time during investigations during 2004 and 57.14% to 100% in 2005. The sensitivity of the SNAP *Giardia* fecal antigen test was comparable, with positive results 50% to 62.5% of the time in 2004 and 85.57% to 100% in 2005. 2nd year veterinary

students evaluated 116 fecal samples from a group of puppies from commercial kennels and recorded that the SNAP *Giardia* fecal antigen test and the $ZnSO_4$ centrifugation technique identified 55 and 45 of the samples as *Giardia* positive, respectively. Twenty-seven participants at a wet lab at the Central Veterinary Conference compared centrifugation versus a passive flotation technique and determined that centrifugation with either 1.18-SG $ZnSO_4$ or 1.27-SG Sheather's sugar solution routinely recovered more eggs and oocysts than the passive Ovassay technique. During this wet lab only once did the Ovassay technique recover the eggs and oocysts of all 6 nematode or coccidian parasites; in contrast, only once did the centrifugation technique fail to recover the eggs and oocysts of all 6 nematode or coccidian parasites. Only six of 27 participants were able to recover and/or identify *Giardia sp.* cysts from a known positive sample using flotation techniques, but all 27 participants had a positive SNAP *Giardia* fecal antigen test.

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An examination of the shedding of cysts and antigen in the feces of laboratory reared Beagle dogs naturally infected with *Giardia*. DWIGHT D. BOWMAN*, JANICE L. LIOTTA, FRANCIS C. DAVIS, KENNETH W. SIMPSON. COLLEGE OF VETERINARY MEDICINE, CORNELL UNIVERSITY, ITHACA, NY.

The availability of 20, laboratory raised and housed dogs that had been identified to be shedding the cysts of *Giardia* in their feces, made it possible to follow the production of cysts and antigen in the feces of the animals over a two-month period. Zinc-sulfate flotation (specific gravity = 1.18) and SNAP (IDEXX Laboratories, Portland, ME) tests were used to initially screen the dogs to identify the 20 animals shedding cysts for inclusion in the study. Fecal examinations did not reveal the presence of other parasites, and bacterial cultures of the feces were negative for *Campylobacter* and *Salmonella*. During the Study, fecal samples from the dogs were examined every Monday, Wednesday, and Friday using both a zinc sulfate flotation (a qualitative "+/-" assessment) and microscope counts of cysts labeled with a commercially available FITC-conjugated monoclonal antibody (Merifluor, Meridian Bioscience Inc., Cincinnati, OH) in 0.1 gram of diluted feces and expressed as cysts per gram dry weight of feces (a quantitative assessment). On Fridays, antigen in the feces was determined qualitatively using IDEXX Snap tests and quantitatively using an antigen detection ELISA (*Giardia* II from TetraTech, Blacksburg, VA) to assess comparative antigen levels shed by each dog. Cysts were not detected in the flotation assay every day in every dog sample examined, but counts using FITC-labeled cyst were consistently high using the dilution cyst count, even in the feces of dogs negative by flotation. Cyst counts per dry weight, normalized by a cube root transformation, remained remarkably consistent throughout the Trial. Antigen was shed regularly by all dogs, and there has been a good correlation between the SNAP tests and the ELISA results. A comparison of the results with the four methods over the 2-month period will be presented.

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An up-date on the prevalence of *Giardia* in "symptomatic" dogs and cats throughout the United States as determined by the IDEXX SNAP® *Giardia* Test. D.D. BOWMAN[1*], J.M. SCARLETT[1], J. GARRETT[2], L. LORENTZEN[2]. [1]COLLEGE OF VETERINARY MEDICINE, CORNELL UNIVERSITY, ITHACA, NY; [2]IDEXX LABORATORIES, INC., WESTBROOK, ME.

Last year we presented on SNAP tests results for *Giardia* antigen on fecal samples from 9,108 symptomatic dogs and 3,071 symptomatic cats. In those samples, there was an overall prevalence of 18.5% antigen positive tests in dogs and 10.8% positive samples amongst the cats. During the past year, we have added 7,006 dog and 1,907 cat samples to the data set. This diagnostic test for detecting the presence of the antigens in the feces of pets makes it possible to look at the national prevalence of this

infection using in-clinic cases because the test is simple to perform in a clinical setting. The results of the test are reproducible, as opposed to flotation and microscopy, because performance does not require superior staff training which minimizes the inherent discrepancies between clinics. The survey again requested that clinics report the results of 20 or more tests performed on dogs or cats with signs of either diarrhea or vomiting. For cats, with the new data the overall prevalence remained almost the same at (10.3%). For dogs, with the new data, the prevalence fell to 15.6%. Breaking the country down into the same regions of West, Midwest, Southeast, and Northeast as utilized by Blagburn et al. (1996, Comp Cont Ed Pract Vet 18:483-509) in their survey of parasites of shelter dogs based on sucrose flotation, there was no significance difference between regions in the case of the cats. However, in the case of dogs, there was a significant increased prevalence of 19.2% in the Northeast and a decreased prevalence of 12.9% in the Southeast compared to the overall prevalence of 15.6% (Pearson Chi-square test, $p<0.001$). Using Mapviewer (Golden Software Inc.), maps have been generated showing the number of samples by state, percentage positive samples by state, and contour maps for the US.

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Evaluation of the association between *Giardia* infection and pruritic skin disease in dogs. A.S. PEREGRINE[1]*, B. SKUBIC-VENGUST[2], J.A. HALL[2], R. REID-SMITH[3], A. LAM[2]. [1]DEPARTMENT OF PATHOBIOLOGY, [2]DEPARTMENT OF CLINICAL STUDIES, [3]DEPARTMENT OF POPULATION MEDICINE, UNIVERSITY OF GUELPH, ONTARIO, CANADA.

Data from children has demonstrated an association between *Giardia* infection and signs of allergy (allergic rhinitis, asthma, atopic dermatitis and urticaria). In order to determine whether or not there is an association between infection with *Giardia* and pruritus in dogs, fecal samples were collected from 71 pruritic dogs with a hypersensitivity dermatosis and 101 clinically normal dogs. Three fecal samples were collected from each animal over 5-7 days and analyzed using a zinc-sulfate concentration technique (ZSCT) for the presence of *Giardia* cysts. In addition, one randomly selected fecal sample from each dog was analyzed with an enzyme-linked immunosorbent assay (ELISA) for *Giardia*-specific antigen (ProSpecT™, *Giardia* Microplate Assay, Alexon Inc.). A questionnaire with emphasis on allergic disease, parasite control and drug use history was administered to all study dog owners. The kappa measure of agreement between ZSCT on three fecal samples and the ELISA on one fecal sample was 0.82, indicating good agreement between tests. Ten of 101 dogs (9.9%) in the clinically normal group tested positive with the ELISA, and *Giardia* cysts were detected in the feces of seven of these antigen-positive dogs. No *Giardia* cysts were found in the dogs that tested negative with the ELISA. None of the dogs in the pruritic group tested positive for *Giardia* by either method. The results indicated a negative association between *Giardia* infection and pruritic skin disease ($p=0.006$). Cephalexin was commonly used in the pruritic group, but not in the control group, and may therefore be an explanatory variable.

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Risk factors for *Cryptosporidium parvum* infection and disease in Ontario dairy calves. L. TROTZ-WILLIAMS[1]*, S.W. MARTIN[1], K.E. LESLIE[1], T. DUFFIELD[1], D.V. NYDAM[2], A.S. PEREGRINE[1]. [1]ONTARIO VETERINARY COLLEGE, GUELPH, ONTARIO, CANADA. [2]COLLEGE OF VETERINARY MEDICINE, CORNELL UNIVERSITY, ITHACA, NY.

Shedding of *Cryptosporidium parvum* oocysts is common in neonatal calves in southwestern Ontario, and is strongly associated with calf diarrhea. In order to identify factors that influence the risk of *C. parvum* infection and diarrhea in dairy calves in this region, fecal specimens were collected in 2003 and 2004 from calves up to 1 month of age on dairy farms with and without histories of calf diarrhea. Specimens were assigned a score to reflect the presence or absence of diarrhea, and were examined for *C. parvum*

oocysts using a sucrose wet mount technique. In addition, questionnaires on farm characteristics and management practices at the farm and calf levels were administered to producers. Statistical regression methods were used for screening of variables and multivariable modelling. Results confirmed that shedding of *C. parvum* oocysts was common in calves in the first month of life; oocysts were detected in fecal samples from 30% of 1089 calves tested in 2004. The presence of concrete flooring in calf housing areas, and the use of soap or detergent when washing calf feeding utensils, were significantly associated ($p<0.05$) with a lower within-herd prevalence of *C. parvum* shedding. Furthermore, calves that were allowed to remain with their dams for more than an hour after birth had a significantly higher risk of diarrhea than those that were separated from their dams within an hour of birth. Calves born in the summer months (May to September) were significantly more likely to shed oocysts than winter-born calves, and calves fed colostrum taken from their own dams had a significantly lower risk of developing diarrhea than those fed colostrum from cows other than their dams.

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Prevalence of *Cryptosporidium* species and genotypes in a 12-month longitudinal study in dairy calves. M. SANTIN*, J.M. TROUT, R. FAYER. ENVIRONMENTAL MICROBIAL SAFETY LABORATORY, ANIMAL AND NATURAL RESOURCES INSTITUTE, AGRICULTURAL RESEARCH SERVICE, UNITED STATES DEPARTMENT OF AGRICULTURE, BELTSVILLE, MD.

Fecal specimens were collected regularly from 30 calves from birth until 12 months of age at a dairy farm in Maryland to determine the prevalence of *Cryptosporidium* species/genotypes. Samples were collected weekly the first 8 weeks after calves were born, biweekly for calves 3 to 5 months of age, and monthly for calves 6 to 12 months of age. Fecal specimens were sieved and subjected to density gradient centrifugation to remove debris and concentrate oocysts. Specimens then were examined by immunofluorescence microscopy and polymerase chain reaction (PCR). All PCR positive specimens were sequenced using the 18S rRNA gene of *Cryptosporidium*. All 30 calves shed *Cryptosporidium* oocysts at some time during the study. Of 630 specimens collected, 163 were infected with *Cryptosporidium* (25.9%). Prevalence was higher in pre-weaned calves (less than 8 weeks of age) (38.3 %) than post-weaned calves (3-12 months of age) (18.2%). Sequence data for 163 PCR-positive specimens identified *Cryptosporidium parvum, C. bovis* and *Cryptosporidium* deer-like genotype. The prevalence of these species and this genotype was age related. *Cryptosporidium parvum,* the only zoonotic species constituted 96.7% of *Cryptosporidium* infections in pre-weaned calves but only 4.2% of *Cryptosporidium* infections in post-weaned calves. The highest prevalence of *Cryptosporidium* was at 2 weeks of age with 28 out of the 30 calves shedding oocysts of *C. parvum*. Our findings clearly demonstrate that the risk of zoonotic infection cannot be determined without molecular characterization to identify the *Cryptosporidium* species/genotype present in fecal specimens. Calves less than 2 months of age are the predominant population infected with *C. parvum* (zoonotic species) and any effort designed to control this infection must be directed primarily at this age group.

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A comparative study of *Cryptosporidium andersoni* and *Giardia* infections in feedlot cattle of Western Australia and Alberta, Canada. BRENDA RALSTON[1]*, R.C.ANDREW THOMPSON[1], DAVE PETHICK[1], TIM MCALLISTER[2], MERLE OLSON[3]. [1] DIVISION OF VETERINARY AND BIOMEDICAL SCIENCES, MURDOCH UNIVERSITY, MURDOCH, WA 6150, AUSTRALIA; [2] AGRICULTURE AND AGRI-FOOD CANADA RESEARCH CENTRE, LETHBRIDGE, ALBERTA, CANADA; [3] BOW VALLEY RESEARCH, CALGARY, ALBERTA, CANADA.

Prevalence, infection pattern and impact on feedlot cattle performance of *Giardia* and *Cryptosporidium* are not well understood. Three studies are being conducted to better understand these factors and their relationship to each other. The first study at Lethbridge, Alberta monitored 300 hd (198 ± 20 kg) of steers collecting 5 fecal samples per animal throughout the feeding period (314d). Prevalence of *Giardia* and *C. andersoni* for this group was 82 and 12%. (Oo)cyst counts of infected steers did not differ significantly (P>0.05) throughout the study. However, the infection rate of *Giardia* decreased on d57 of the study. There was no significant difference in average daily gain (ADG) between *Giardia* or *C. andersoni* infected and non-infected steers. The second study at Lethbridge monitored 542 steers (243 kg) sampling occurring every 21 days. Preliminary results (3 samples) indicate a lower prevalence of both Giardia (25%) and C. andersoni (3%) compared to the first study. Currently there is no significant difference in average daily gain (ADG) between *Giardia* or *C. andersoni* infected and non-infected steers. The third study conducted in a feedlot in Western Australia involves 244 steers (486 kg). Overall prevalence of *Giardia* and *C. andersoni* to date are 7 and 8% respectively. Performance data indicates no significant difference in ADG between *Giardia* or *C. andersoni* infected and non-infected steers. However, a subset of the group comprised of steers averaging 110.7 kg less than their contemporaries had a prevalence of 17 and 26% for *Giardia* and *C. andersoni* respectively and the *C. andersoni* infected steers had a lower ADG than their contemporaries (P=0.07). Preliminary results from these three trials demonstrate the variability in prevalence and impact on performance of *Giardia* and *C. andersoni* infections between groups of cattle and the importance of further research to elucidate the factors involved in this variation.

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Equine Cyathostomins: Secretory and inflammatory status in the ventral colon of naturally infected and maximally treated ponies. T.R KLEI*, R. McCONNICO, S.R.CHIRGWIN, J.D. HUBERT, S.U.COLEMAN. SCHOOL OF VETERINARY MEDICINE, LOUISIANA STATE UNIVERSITY, Baton Rouge, LA.

The current study was conducted to further test the hypothesis that the steady state cyathostomin infection causes a mucosal inflammatory response which alters epithelium secretory function. Twelve mature mixed breed ponies were allowed to graze a cyathostomin contaminated pasture for one year. Ponies were ranked by EPG levels, placed in one of two groups and housed in stalls. One group (Tx) was treated with OBZ at 20 mg/kg for 5 days, followed by one dose of IVM at 200ug/kg. The second group (NTx) remained untreated. Necropies were preformed on ponies in group NTx 4 wks after being stalled. Necropsies of Tx animals were preformed 7 wks after treatment. Mucosal parasite burdens determined by digestion, were high in the NTx group (mean 36, 403). While there was a 86% reduction in the Tx group significant numbers of parasites remained (mean 5,168). Histologic samples indicate that eosinophil and mast cell numbers were increased in the mucosa of NTx individuals. Right ventral colonic LN were removed, pooled and mRNA levels of IL-1,4,5,6,8 and IFN gamma and TNF alpha measured by real time RT-PCR. IL-5 and IFN gamma levels were elevated in both groups but more so in Tx individuals. Chloride secretion as reflected by short circuit current in an Ussing chamber suggests that secretory activity was increased in the Tx group. These observations suggest that while moderate to heavy infection of cyathostomins increase the numbers of inflammatory cells it is not sufficient to increase secretory activity of the epithelium. However, the activation of hypobiotic EL3 may alter this activity.

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Controlling gastrointestinal nematode infection in goats using sun-dried forage high in condensed tannins. T.H. TERRILL[1*], G.S. DYKES[1], S.A. SHAIK[1], J.E. MILLER[2], B. KOUAKOU[1], G. KANNAN[1], J.M. BURKE[3], R. M. KAPLAN[4], J.A. MOSJIDIS[5]. [1]FORT VALLEY STATE UNIVERSITY, FORT VALLEY, GA, [2]LOUISIANA STATE UNIVERSITY, BATON ROUGE, LA, [3]USDA, ARS, DBSFRC, BOONEVILLE, AR, [4]THE UNIVERSITY OF GEORGIA, ATHENS, GA, [5]AUBURN UNIVERSITY, AUBURN, AL.

Gastrointestinal nematode (GIN) parasitism is the greatest threat to economic sheep and goat production in the southern USA, and there is widespread prevalence of GIN resistance to anthelmintics in this region. A natural control alternative is feeding hay of sericea lespedeza (*Lespedeza cuneata*), a perennial warm-season legume high in condensed tannins. Two confinement trials were completed with Spanish/Boer/Kiko cross yearling bucks given unground (Trial 1) and ground (Trial 2) hay of SL and bermudagrass (*Cynodon dactylon*). The bucks were naturally infected, supplemented by a trickle infection (*Haemonchus contortus*) in trial 1. Goats were fed hay treatments at 75% of daily intake, with 25% supplement in trial 1. In trial 2, the hay treatments were combinations of SL (0, 25, 50, and 75%) and BG (75, 50, 25, and 0%), with all animals receiving 25% supplement. Goats were fed the treatment

diets for seven and six weeks in trials 1 and 2, respectively. Fecal and blood samples were collected from individual animals weekly to determine FEC and packed cell volume (PCV), respectively. Adult worms were recovered from the abomasum and small intestine of the goats at the end of each trial. Feeding SL hay to goats significantly ($P < 0.05$) reduced FEC and increased PCV compared with BG hay. Effects at 50 and 75% of the diet were similar, with some positive effect at 25%. A lower percentage of ova in feces from SL-fed goats developed into infective (L3) larvae. There was a direct effect of SL hay on adult GIN, with significantly ($P < 0.01$) lower numbers of both abomasal (*H. contortus*, *Teladorsagia circumcincta*) and small intestinal (*Trichostrongylus colubriformis*) nematodes compared with goats fed BG hay. Feeding SL hay to goats is an effective means of controlling parasitic nematodes and may be a potential supplement/replacement for chemical anthelmintics.

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Efficacy evaluation of macrocyclic lactone pour-ons under field conditions. K.B. HART[1]*, [2]D. H. BLISS, [1]SCHOOL OF VETERINARY MEDICINE, UNIVERSITY OF ILLINOIS, URBANA, IL, [2]MIDAMERICA AG RESEARCH, VERONA, WI.

Six field trials were conducted with two separate beef herds at the University of Illinois Dixon Springs Agriculture Research Center located in southern Illinois to investigate whether exclusive use of eprinomectin (Ivomec® Eprinex™ – Merial) pour-on over the past few years has led to the development of parasite resistance to eprinomectin and moxidectin. A standard Fecal Egg Count Reduction Test (FECRT) was conducted on pre-treatment and post-treatment fecal samples. Utilizing a randomized block design with pretreatment egg counts used as a blocking factor, six trials were completed as follows: 1 and 4) Eprinomectin and moxidectin (Cydectin® - Ft. Dodge Animal Health) pour-on, respectively, treated calves versus non-medicated control calves, 2 and 5) Fenbendazole (Safe-Guard™/ Panacur® – Intervet, Inc.) treated calves versus non-medicated control calves and 3 and 6) Eprinomectin and moxidectin, respectively, treated calves versus fenbendazole treated calves. Fecal samples were analyzed at two separate labs for Trials 1, 2 and 3 using the Modified Wisconsin Sugar Flotation test and the Sheather's Sugar Solution Fecal Flotation Technique. Fecal samples from Trials 4, 5 and 6 were analyzed only using the Modified Wisconsin Sugar Flotation Technique. The results were comparable and demonstrated significant reductions in the efficacy of eprinomectin (84.8% efficacy in Trial 1, 5.5% efficacy in Trial 3) and moxidectin (74.7% efficacy in Trial 4, 0% efficacy in Trial 6) had occurred indicating parasite resistance to eprinomectin and cross resistance to moxidectin had developed in these two beef herds. Fenbendazole was used as a positive control dewormer and demonstrated a high degree of efficacy (97.5% efficacy in Trial 3, 99.9% efficacy in Trial 6) against the same parasites demonstrating resistance to eprinomectin and moxidectin.

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Efficacy of fenbendazole and milbemycin oxime for treating baboons infected with *Trichuris trichiura.* MASON V. REICHARD[1]*, ROMAN R. WOLF[2], DAVID W. CAREY[2], JENNIFER J. GARRETT[3], HEATHER A. BRISCOE[1]. [1]DEPARTMENT OF VETERINARY PATHOBIOLOGY, CENTER FOR VETERINARY HEALTH SCIENCES, OKLAHOMA STATE UNIVERSITY, STILLWATER, OK, [2]ANIMAL RESOURCES, UNIVERSITY OF OKLAHOMA HEALTH SCIENCES CENTER, OKLAHOMA CITY, OK, AND [3]DEPARTMENT OF BIOLOGY, TARLETON STATE UNIVERSITY, STEPHENVILLE, TX.

The efficacy of fenbendazole (FBZ) and milbemycin oxime (MO) for treating baboons infected with *Trichuris trichiura* was evaluated by using fecal egg count reduction tests. Six baboons, each singly housed at the University of Oklahoma Health Sciences Center and known to be infected with *T. trichiura,*

were assigned to one of two treatment groups. There were three experimental animals in each treatment group and each orally received therapeutic doses of either FBZ (group A) or MO (group B) mixed with their food. Within 6 days of treatment with FBZ, there was a 100% reduction in the number of *T. trichiura* eggs shed per gram of feces from all three baboons in group A. Fecal egg counts remained negative for *T. trichiura* from group A baboons 60 days post-treatment. Conversely, comparisons of the number of *T. trichiura* eggs shed per gram of feces pre- and post-treatment from group B baboons that received MO showed that there was not a significant reduction. The number of *T. trichiura* eggs per gram of feces increased by 134.1% post-treatment for one of the baboons in group B whereas reductions in egg counts of only 4.6% and 29.0% were observed from the other two baboons that received MO. During the same time period of pre- and post-treatments fecal eggs count for *T. trichiura* from baboons in groups A and B, a control baboon that did not receive anthelmintic treatment had a reduction of 24.8% in the number of *T. trichiura* eggs shed per gram of feces. The results of this study indicate that FBZ was extremely effective for treating baboons infected with *T. trichiura* whereas treatment with MO had little to no effect.

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Current status of canine heartworm disease in Korea and its potential public health importance. SUNGSHIK SHIN[1]*, SHINYOUNG KANG[1], JUNGKEE KWON[2]. [1]COLLEGE OF VETERINARY MEDICINE, CHONNAM NATIONAL UNIVERSITY, GWANGJU, KOREA, AND [2]COLLEGE OF VETERINARY MEDICINE, CHONBUK NATIONAL UNIVERSITY, KOREA.

Since canine dirofilariasis in South Korea was first reported in 1962, the causative parasite, *Dirofilaria immitis*, is continuing to spread within the country. A survey of canine dirofilariasis in five areas of South Korea in 1994, for instance, revealed that 28.3% of 127 dogs were infected with *D. immitis*. In 2001, however, the prevalence of heartworm infection among 165 dogs in three breeding farms in the vicinity of Seoul, Korea was 50.3%. Between 2004 and 2005, another survey of can! ine dirofilariasis in Jindo Island showed that 254 of 365 dogs (69.6%) were infected with *D. immitis*. Moreover, a highly endemic area of canine dirofilariasis was yet located in Asan where 26 of 30 dogs (86.7%) were infected. Presence of *D. immitis* was also documented in other animals such as cats, raccoon dogs, harbor seals, and jaguars in Korea. An echocardiographic study compared with necropsy findings performed on 12 Jindo dogs naturally infected with heartworms indicated that the adult heartworms were located mostly in the terminal branches of the pulmonary arteries rather than in the heart when the host was alive, but the worms appeared to move toward the right ventricle shortly after the he! art of the infected dogs stopped beating. Clinical signs of canine dirofilariasis did not necessarily correlate with the number of adult worms. Several field trials have been performed in Korea using commercial prophylactic agents with avermectin derivatives as the active ingredient which included ivermectin, selamectin and moxidectin. A considerable variation in the accuracy of 9 commercially available diagnostic kits was noticed. As the incidence of canine dirofilariasis appears to be increasing in Korea, this entity represents an important zoonosis and indicates a need for familiarity with the pathologic features of the lesions and the morphologic characteristics of the worm.

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Results and realistic implications of the 2004 AHS-Merial heartworm survey. JORGE GUERRERO[1]*, CT NELSON[2], DS CARITHERS[3]. [1]DEPT OF PATHOBIOLOGY, SCHOOL OF VETERINARY MEDICINE, UNIVERSITY OF PENNSYLVANIA, PHILADELPHIA, PA, [2]AMERICAN HEARTWORM SOCIETY (PRESIDENT), ANIMAL MEDICAL CENTER, ANNISTON, AL AND [3]MERIAL LIMITED, DULUTH, GA.

In 2005, a heartworm survey was sent out to over 25,000 veterinary practices by the American Heartworm Society, in cooperation with Merial. Responses were received from more than 12,000 veterinary clinics, and based upon these responses, more than 250,000 dogs in the US tested positive for heartworms in 2004. These numbers were up slightly from 244,000 positive dogs noted in a similar survey, with a similar response rate, performed in 2001. Although response rate was high, the survey data accounts for less than one-half of the total veterinary clinics in the United States. The American Veterinary Medical Association (AVMA) reported that, of the 61.6 million dogs in the US, 51.5 million (83.6%) are seen by veterinarians at least once a year.[1] Furthermore, according to polls by Gallup[2] and B&R Tracking[3], heartworm disease preventives are only dispensed to ~58% of dog-owning households, a 8% decline from the 66% dispensing rate reported in 1998. The information on non-reporting clinics in the 2004 survey, AVMA data on dogs never seen by veterinarians, and decreased preventive dispensing, would indicate that heartworm disease is likely a much more significant issue than the AHS/Merial heartworm survey numbers indicate. Both the 2001 and 2004 heartworm surveys were performed to increase awareness and provide a tool for client education. They were intended to illustrate the need for heartworm disease prevention for pet owners and veterinary staff. While useful as an education tool, these surveys underestimate the prevalence of canine heartworm disease. These survey results demonstrate a need for continued, consistent and aggressive heartworm education. Future practitioners and veterinary technicians, current practitioners, clinic staff, and pet owners, all need education and awareness to decrease the real number of heartworm positive dogs in the US.

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Data from the 1992-2002 annual heartworm surveys for Ontario, Canada and effectiveness of preventive medication. OWEN SLOCOMBE*, DEPARTMENT OF PATHOBIOLOGY, ONTARIO VETERINARY COLLEGE, UNIVERSITY OF GUELPH, GUELPH, ONTARIO, CANADA .

In Canada, annual questionnaire surveys for *Dirofilaria immitis* in dogs were conducted since 1976. Data from a small section of the 1992 - 2002 surveys is presented for the province of Ontario, the primary focus of heartworm (HW) infection in Canada. During that period, cases reported for Canada and Ontario ranged from 327 - 812 and 240 - 676 respectively. Other information presented here is only for Ontario with ranges for the 1992 - 2002 period. The response rate to the questionnaires was 60 - 73%. Number of blood tests reported and prevalence estimated were 255,360 - 289,289 and 0.09% - 0.25% respectively. Exceptionally few dogs (range 20 - 84) that had preventive medication tested HW positive. Most positive dogs (range 233 - 612) never had medication. HW cases/20,000 tests for dogs that had medication ranged from 1.9 - 7.4 and for dogs that did not from 95.7 - 203.0. From 1993 onwards, there was a gradual trend upwards in numbers in all those parameters with a marked increase in 1996 - 1998, a marked decline in 2000 and subsequently a gradual decline to 2002. For about 53% of HW dogs that had medication, the owners admitted to missing medicating or improper dosage. For the other 47%, principal reasons were the dogs the previous year and prior to medication were (1) diagnosed with HW and (a) not given an adulticide, or (b) given an adulticide but with inadequate or no determination of its effectiveness, (2) tested only for microfilariae and negative, but the next year tested for antigen and positive, (3) not tested or tested negative (type of test unknown) and the next year tested at another clinic and positive. Owner non-compliance with use of HW preventive medication and product failure were minor issues during 1992-2002.

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Clinical and pathologic reactions in dogs of heartworm extract-induced shock. M.A.G. BARBOSA[1]*, R.A. MOTA[1], L. C. ALVES, L.C, F. L. C. BRITO[2], E. R. ALBUQUERQUE[1], C. R. A. NASCIMENTO[1], M. N. TEIXEIRA[1]; F. C. L. MAIA[1], D. SOUZA[3], M. A. G. FAUSTINO[1]. [1]DEPARTMENT OF VETERINARY MEDICINE-UNIVERSIDADE FEDERAL RURAL DE PERNAMBUCO , BRAZIL, [2] FACULDADE DE CIENCIAS AGRARIAS -UNIVERSIDADE ESTADUAL DE SÃO PAULO , BRAZIL.

To evauate the clinical signs and shock-like reaction in dogs of heartworm (HW) extract-induced shock, studies were performed in two groups of mixed-breed male dogs containing six (Experimental Group) and two (Control Group) animals which were administered IV a crude, whole-body extract of female heartworms or saline solution respectively. The clinical signs and shock-like reaction as well as changes in clinical parameters such as electrocardiographic record, RBC and WBC counts, serum alanine amino transferase, aspartate amino transferase, creatine phosphokinase and also urea were monitored . Changes in clinical, hematological findings and clinical chemistry were examined from before treatment to 24 hours after treatment in dogs given HW extract and also saline solution. The results had been analyzed by Wilcoxon and Mann-Whitney tests for clinical hematological, clinical chemistry and by the Fischer test to electrocardiographic values respectively. Shock with clinical signs such as the pale color of the visible mucous membranes, respiratory disorders, vocalization and defecation were observed in some of experimental group of dogs. It was observed a significative difference not only between the lynfocytes count and CPK values but also with RBC and eosinophil count. These resuts showed the clinical, hematologic, and clinical chemistry findings which happen after the drug administration in dogs with HW infection.

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Effects of ivermectin and doxycycline administered alone, together, or together plus melarsomine against adult *D. immitis* in dogs with induced infections: Clinical observations. M. T. DZIMIANSKI[1]*, J. W. MCCALL[1], R. ROBERTS[2], S. D. MCCALL[3], L. H. KRAMER[4], C. GENCHI[5], J. GUERRERO[6], A. MANSOUR[3], N. SUPAKORNDEJ[3], P. SUPAKORNDEJ[1]. [1]DEPARTMENT OF INFECTIOUS DISEASES, COLLEGE OF VETERINARY MEDICINE, UNIVERSITY OF GEORGIA, ATHENS, GA; [2]DEPARTMENT OF ANATOMY AND RADIOLOGY, COLLEGE OF VETERINARY MEDICINE, UNIVERSITY OF GEORGIA, ATHENS, GA; [3]TRS LABS, INC., ATHENS, GA; [4]DEPARTMENT OF ANIMAL PRODUCTION, COLLEGE OF VETERINARY MEDICINE, UNIVERSITY OF PARMA, ITALY; [5]DEPARTMENT OF VETERINARY PATHOLOGY AND PARASITOLOGY, UNIVERSITY OF MILAN, ITALY; [6]DEPARTMENT OF PATHOBIOLOGY, UNIVERSITY OF PENNSYLVANIA, PHILADELPHIA, PA.

Laboratory and clinical studies have demonstrated that endosymbiotic *Wolbachia* are an important factor in the pathogenesis of filarial disease in man. Tetracycline antibiotics have been shown to inhibit filarial larval development and embryogenesis in adult filariae. Data also suggests that depletion of these intracellular bacteria with antibiotics may reduce the inflammatory reactions following antifilarial treatment. To determine if depletion of *Wolbachia* in *Dirofilaria immitis* by antibiotic treatment would provide clinical benefit, dogs with patent infections of heartworm were treated with doxycycline (DOXY), ivermectin (IVM), and melarsomine (MEL) either singly or in various combinations (DOXY+IVM and DOXY+IVM+MEL). Details of the treatment are given in the preceding presentation (McCall et al.). During this study, clinical assessments (physical examination, radiography, clinical chemistries and CBC, and urinalysis) were made on all dogs at the onset of the treatment phase and at 12, 24, and 36 weeks. Additional assessments were made at 28 and 32 weeks for dogs given Melarsomine.

Necropsy evaluations were made at 36 weeks. Clinical findings of significance included the following: 1. eosinophilia was found in most dogs treated with either Dox or Ivm alone at 12 weeks following a precipitous decline in microfilaremia, 2. only dogs that received IVM did not have elevations in serum alanine aminotransferase (ALT) concentrations, and 3. treatment with MEL had no apparent effect on serum clinical chemistries. At necropsy, gross pathological lesions were most severe in dogs treated only with MEL. In contrast, dogs given the combination DOXY+IVM+MEL had the fewest lung lesions with 3 of 5 dogs having no grossly visible lesions. The clinical relevance of the data will be discussed.

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Effects of ivermectin and doxycycline administered alone, together, or together plus melarsomine against adult *D. immitis* in dogs with induced infections: Parasitological data. J. W. MCCALL[1]*, S. D. MCCALL[2], L. H. KRAMER[3], N. SUPAKORNDEJ[2], A. MANSOUR[2], P. SUPAKORNDEJ[1], M. T. DZIMIANSKI[1], R. ROBERTS[4], C. GENCHI[5], J. GUERRERO[6]. [1]DEPARTMENT OF INFECTIOUS DISEASES, COLLEGE OF VETERINARY MEDICINE, UNIVERSITY OF GEORGIA, ATHENS, GA; [2]TRS LABS, INC., ATHENS, GA; [3]DEPARTMENT OF ANIMAL PRODUCTION, COLLEGE OF VETERINARY MEDICINE, UNIVERSITY OF PARMA, ITALY; [4]DEPARTMENT OF ANATOMY AND RADIOLOGY, COLLEGE OF VETERINARY MEDICINE, UNIVERSITY OF GEORGIA, ATHENS, GA; [5]DEPARTMENT OF VETERINARY PATHOLOGY AND PARASITOLOGY, UNIVERSITY OF MILAN, ITALY; [6]DEPARTMENT OF PATHOBIOLOGY, UNIVERSITY OF PENNSYLVANIA, PHILADELPHIA, PA.

Thirty beagles were infected with 7 adult male and 9 adult female heartworms by IV transplantation. Six weeks later, dogs were ranked by Mf and randomly allocated to 6 groups of 5 dogs (Day 0). Group 1 received Heartgard Plus (6 mcg/kg) (IVM) weekly for 9 months. Group 2 received doxycycline (10 mcg/kg/day) (DOXY) orally during weeks 1-6, 10-11, 16-17, 22-25, and 28-33. Groups 3 and 5 received both IVM and DOXY. At Week 24, Groups 3 and 4 received an IM injection of melarsomine (2.5 mg/kg) (MEL), followed one month later by 2 injections 24 hours apart. Group 6 was not treated. Whole blood/serum samples were collected for Mf/Ag testing, immunologic evaluation, and molecular biology procedures before infection, before IVM and DOXY dosing, and at Weeks 6, 10, 12, 16 (Mf/Ag only), 18 (Mf/Ag only), 24, 26 (Mf/Ag only), 28, 32, and 36. Radiography, physical examination, hematology/clinical chemistry, and urinalysis were done before infection (RAD only), before Day 0, and at Weeks 12, 24, 28 (Groups 3&4), 32 (Groups 3&4), and 36. At 36 weeks, dogs were humanely euthanized and necropsied for worm recovery, collection of lung, liver, kidney, and spleen samples, and later examination of these tissues by immunohistochemistry and conventional histologic methods. Heartworms were weighed and prepared for examination by immunohistology, conventional microscopy/TEM, and molecular biology techniques. All 10 dogs treated with both IVM and DOXY were amicrofilaremic after Week 9. Microfilaremias gradually decreased in dogs treated with IVM or DOXY, but most had a few microfilariae at necropsy. Antigen scores gradually decreased with IVM plus DOXY and after MEL (1.8-2.2), while scores for IVM or DOXY alone were similar to controls (4.0-4.2 at necropsy). Percent reduction of adult worms for IVM, DOXY, IVM/DOXY/MEL, MEL, and IVM/DOXY treatment were 20.3, 8.7, 92.8, 100, and 78.3.

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Anthelmintic resistance in New Zealand: a canary in a coal mine for US producers?

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A recent national survey of anthelmintic resistance prevalence and associated practices in New Zealand revealed surprising levels of anthelmintic inefficacy. This presentation will discuss those results, likely causative factors and sustainability strategies. This information will be more widely relevant as the anthelmintic resistance conversation gets louder in the United States. Fecal Egg Count Reduction Tests (FECRT) were performed on 80 sheep and 62 beef properties. Oral ivermectin failed to reduce FEC by 95% on 25% of surveyed sheep properties, albendazole on 41% and levamisole on 24%. An albendazole/levamisole combination failed on 8% of properties. The parasite most commonly implicated in sheep was *Teladorsagia circumcincta.* Ivermectin failed on over 90% of beef properties, albendazole on 76% and levamisole on 8%. The major parasite involved was *Cooperia oncophera.* The gravity of these results and their contributory management practices has shaken stakeholders in the NZ livestock industry. Producer preference for formulations featuring application convenience and long activity in the face of high pasture larval challenge is a likely contributing factor, especially in cattle. Discussion with US colleagues suggests that parallel attitudes and practices in US systems are creating the same worrisome results. Progressive NZ producers and their advisors are integrating combination anthelmintics and refugia as sustainability strategies. Combination anthelmintics have been available for use in sheep and cattle in NZ for several years. Oral BZ/Levamisole combinations have been licensed since 1990. Macrocyclic Lactone (ML)/BZ/LEV combinations have been available to sheep producers since 2001. Two years ago an abamectin/levamisole combination pour-on was registered for use in cattle. In the absence of promising new molecules combinations will play a vital future role in New Zealand parasite management. A recent national survey of anthelmintic resistance prevalence and associated practices in New Zealand revealed surprising levels of anthelmintic inefficacy. This presentation will discuss those results, likely causative factors and sustainability strategies. This information will be more widely relevant as the anthelmintic resistance conversation gets louder in the United States. Fecal Egg Count Reduction Tests (FECRT) were performed on 80 sheep and 62 beef properties. Oral ivermectin failed to reduce FEC by 95% on 25% of surveyed sheep properties, albendazole on 41% and levamisole on 24%. An albendazole/levamisole combination failed on 8% of properties. The parasite most commonly implicated in sheep was *Teladorsagia circumcincta.* Ivermectin failed on over 90% of beef properties, albendazole on 76% and levamisole on 8%. The major parasite involved was *Cooperia oncophera.* The gravity of these results and their contributory management practices has shaken stakeholders in the NZ livestock industry. Producer preference for formulations featuring application convenience and long activity in the face of high pasture larval challenge is a likely contributing factor, especially in cattle. Discussion with US colleagues suggests that parallel attitudes and practices in US systems are creating the same worrisome results. Progressive NZ producers and their advisors are integrating combination anthelmintics and refugia as sustainability strategies. Combination anthelmintics have been available for use in sheep and cattle in NZ for several years. Oral BZ/Levamisole combinations have been licensed since 1990. In recent years Macrocyclic Lactone (ML)/BZ/LEV combinations have become available to sheep producers. Two years ago an abamectin/levamisole combination pour-on was registered for use in cattle. In the absence of promising new molecules combinations will play a vital future role in New Zealand parasite management.

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Macrocyclic lactone-resistant *Oxyuris equi*: Anecdote or emerging problem?

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Several locations have recently reported equine pinworm (*Oxyuris equi*) infections that were refractory to repeated treatment with ivermectin. Definitive investigations have been hampered by a lack of parasitologic evidence, and by the low sensitivity of diagnostic techniques for pinworms. During 2005 in Tennessee, perianal scrapings were examined to demonstrate patent *Oxyuris* infections in 4 and 4 horses at Farms A and B, respectively, despite historical ivermectin treatments within the prepatent period. After diagnosis, all infected horses were treated orally with Zimecterin® paste (ivermectin, 0.2 mg/kg). Approximately 14 days later, each horse was treated again with oral pyrantel pamoate paste (6.6 mg/kg), and feces were collected for 72 hours post-treatment. Feces were washed over a #10 (2 mm aperture) sieve, and adult pinworms were recovered, counted, and preserved in 10% formalin. At Farm A, 1 of 4 horses passed 2 adult *Oxyuris*; at Farm B, 2 of 4 horses passed 2 and 25 adult specimens. Some mature pinworms obviously survived ivermectin treatment in both herds, but it is unclear whether these represented anthelmintic-resistant populations or merely indicated incomplete efficacy of the macrocyclic lactones. (In the ivermectin and moxidectin FOIs, efficacy claims against adult *Oxyuris equi* were 90-100%.) It is interesting to speculate that the biology of *Oxyuris* might also be changing, in addition to its anthelmintic susceptibility profile. Acquired immunity typically restricts adult pinworm infection to horses $\leq$2 years of age. However, most reports of inefficacy have involved mature horses, and infected subjects in TN ranged from 5 to 25 (mean = 14.0) years of age.

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Continued evaluation of combination anthelmintics treatments in a commercial cattle population where anthelmintic resistant parasites had been observed the previous years.

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More than 4,300 calves weighing 70-100 kg were purchased in the southeastern US during the winter of 2004-2005, and then were placed in either a conditioning yard for 40-60 days prior to pasturing, or were taken directly to pasture. All calves received doramectin when purchased. Calves placed in a conditioning yard were further treated with moxidectin prior to moving to pasture. Calves that went directly to pasture were treated once or twice with either ivermectin plus clorsulon or doramectin. Between May 5-8 the animals were reassembled, sorted by size and sex, treated with ivermectin plus clorsulon and levamisole (only ½ the animals received the levamisole) and shipped to Wisconsin. In Wisconsin all animals, except for one pasture, were intensively rotationally grazed through the summer and fall of 2005. All fecal samples were randomly taken from the pastures and reported as EPG. On pastures **SH**, 690 heifers averaging 313 kg at turnout were treated with Ivomec and levamisole on 9 June and treated on 27 July with Cydectin and levamisole and on September 15 with Ivomec and levamisole. Egg counts were: 20 samples on 17 Aug = .94, 17 samples on 29 Sept=0, 9 samples on 31 October = 0 and 20 samples on15 December =3.13. On pastures **SS**, 603 steers averaging approximately 155 kg at turnout were retreated with Ivomec and levamisole on 9 June, retreated with Cydectin and levamisole on 20 July. On 09 September the EPG =35.4. After reviewing these results the owner decided to redeworm all groups and not run more fecal samples at this time. Fenbendazole in the feed was used because the animals were not near the corral. Animals averaged 600 lbs but owner decided to use a 700 lbs dosage from 16 - 21 September. Egg counts were: 15 samples on 4 October = .68; 11samples on 31 October=1.9;19 samples on 15 Dec. = 3.6. On **LH** pastures, 640 heifers averaging approximately 211 kg at turnout were treated with Ivomec and levamisole on 17 June and retreated on 26 July with Cydectin

and levamisole. From September 22-28, retreated with fenbendazole in the feed at a dose of approximately 100 # more than animal average weight. Egg counts were: 16 samples on 12 October = .56; 8 samples on 31 October = 2.83; 20 samples on 15 Dec.=3.06. On **LS** pastures, 720 steers averaging approximately 218 kg at turnout were treated on 15 June and 28 July with Cydectin and levamisole; September 22-28, because the animals were not near the corral, fenbendazole was put in the feed. Egg counts were: 17 samples on 12 Oct= .7; 10 samples 31 Oct=1.2;19 samples on 15 Dec=3.6. On **MH** pastures, 703 heifers averaging approximately 187 kg at turnout were treated with Ivomec and levamisole on 10 June; treated with Cydectin and levamisole on 18 July; September 22-28, because the animals were not near the corral, fenbendazole was placed in the feed at a dose of approximately 100 pounds more than the animals' average weight. Egg counts were; 15 samples on 12 Oct= 1.2; 10 samples on 31 Oct= 0; 20 samples Dec15 =2.38. On **MS** pastures, 875 steers averaged approximately 188 kg at turnout. On 14 June the first ½ were treated with Eprinex and levamisole; because of rain the last ½ were treated with Ivomec and levamisole; treated on 01 Aug and 20 Sept. with Cydectin and levamisole. Egg counts were; 15 samples on 04 Oct=0; 11 samples on 31 Oct = 0. Animals sold in early Nov; no samples taken. On pasture **G** (not intensively rotationally grazed), 158 animals averaging 120 kg at turnout were treated on 13 June with Ivomec and levamisole; 19 July with Cydectin and levamisole; 16 September with Ivomec and levamisole. EPG were: 15 samples on 6 Sep = 50.6; 13 samples on 29 Sep= 0; 8 samples on 31 Oct=9. Cattle removed the first week of Nov.

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Resistance to Gastrointestinal nematodes of cattle: Identification of genomic regions affecting resistance and potential mechanisms. LOUIS C. GASBARRE*, TAD S. SONSTEGARD, ROBERT LI, RICARDO ARAUJO, CURTIS P. VAN TASSELL. BOVINE FUNCTIONAL GENOMICS LAB, ARS, USDA, BELTSVILLE, MD.

Gastrointestinal nematode infections remain a major economic drain on the efficient raising of cattle throughout the world. The recent demonstrations of the appearance of drug resistance in these parasites underscores the problems associated with a complete reliance on anthelmintics to control economic loss. A number of years ago we began the development of a resource population of Angus cattle to map and identify important bovine genes that affect resistance to these parasites. The pedigree of this population encompasses over 600 animals and approximately 500 of these have been completely characterized for their resistance to natural challenge by the parasites. Over the period of this work, we have seen the segregation of resistant and susceptible phenotypes and can now predict with about 80% accuracy the phenotype of calves prior to actual testing. The development of these animals has allowed us to begin to identify the areas of the genome that contain genes important in determining the resistance phenotype of individual animals, and also provided us with animals in which we can begin to determine the actual mechanisms of resistance, To date, we have identified 8 regions on 6 chromosomes that affect resistance to the parasites. In terms of mechanism(s) of resistance, it appears that resistant animals are more efficient in the generation and maintenance of inflammatory responses at the actual site of infection, i.e. the gut mucosa. The pending release of the bovine genome sequence and associated SNP HapMap will greatly facilitate the identification of the actual structural genetic polymorphisms that render animals resistant or susceptible to the parasites.

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Ivermectin and moxidectin cause constitutive and induced overexpression of different P-glycoproteins in resistant *Haemonchus contortus*. ALAIN ROULET, ROGER PRICHARD*. INSTITUTE OF PARASITOLOGY, McGILL UNIVERSITY, MONTREAL, CANADA.

We have previously reported evidence that ivermectin and moxidectin cause genetic selection on P-glycoprotein A (PgpA) and overexpression of PgpA in *Haemonchus contortus*. Using realtime PCR, we have measured the levels of mRNA for six P-glycoproteins in four strains/isolates of *H. contortus*. These isolates were an unselected strain (PF), two laboratory strains, derived from the PF strain, selected with ivermectin (IVF) or moxidectin (MOF), and a field mutli-drug, including macrocyclic lactone, resistant strain (Wallangra). Ivermectin selection/resistance resulted in an increase in constitutive expression of several of the Pgps, while moxidectin selection also resulted in an increase in constitutive expression of some of the Pgps; but generally to a lesser extent than ivermectin.
The induction of overexpression, by these macrocyclic lactones, was also examined in these Pgps in the field strain that was strongly ivermectin resistant and moderately resistant to moxidectin. Ivermectin induced further overexpression, 24 hours after treatment of infected sheep, in four of the *H. contortus* Pgps, while moxidectin induced further overexpression in one of the same Pgps, and another Pgp not induced by ivermectin treatment. These results show that constitutive and induced overexpression of some P-glycoproteins is associated with ivermectin resistance and also with moxidectin selection. However, the constitutive and induced P-glycoprotein profiles are different between ivermectin and moxidectin. Supported by NSERC, CHPI and Fort Dodge Animal Health.

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Application of polymerase chain reaction (PCR) and drug-sensitivity testing to compare species composition and anti-coccidial drug resistance in *Eimeria* isolated from vaccine- and coccidiostat-utilizing poultry operations. M.C. JENKINS[1], S. KLOPP[2], G. WILKINS[1], K. MISKA[1]. [1]ANIMAL PARASITIC DISEASES LABORATORY, ARS, USDA, BELTSVILLE, MD. [2]TOWNSENDS, INC., GEORGETOWN, DE.

The purpose of this study was to compare drug resistance and species composition of *Eimeria* isolated from two types of poultry operations that differed in the means of controlling avian coccidiosis. The number of species present in vaccine-control facilities was greater than in drug-control operations, with all 6 species present in the former, and 3-4 species present in the latter. *Eimeria* propagated from drug-utilizing operations displayed greater anti-coccidial resistance compared to *Eimeria* recovered from farms using a live oocyst vaccine. These results suggest that conventional drug treatment leads to lower *Eimeria* species diversity coincident with slightly greater drug resistance. An improved ITS1-rDNA PCR method was used to follow the expansion of drug-resistant *Eimeria* in chickens raised in the presence or absence of specific anti-coccidial compounds. Drug-resistant coccidia rapidly dominated the population of *Eimeria* spp. oocysts, which has relevance to controlling avian coccidiosis with drugs and live oocyst vaccines.

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Confirmation of ivermectin resistance in a purportedly resistant Canadian isolate of *Parascaris equorum* in foals
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COLLEGE, UNIVERSITY OF GUELPH, GUELPH, ONTARIO, CANADA, [4]MERIAL LIMITED, DULUTH, GEORGIA.

Ivermectin is the most commonly used anthelmintic in foals due to its broad spectrum of activity and excellent safety record. Resistance to benzimidazole anthelmintics in equine cyathostome nematodes has been recognized as an important problem for many years, but until the recent reports of suspected resistance in *Parascaris equorum*, ivermectin resistance had not been diagnosed in any equine parasite. *Parascaris* fecal egg counts (FEC) do not correlate with worm counts in foals; therefore we conducted a controlled efficacy study to investigate the possible occurrence of ivermectin resistance in *P. equorum*. In this study, 11 foals were infected orally with 500 eggs of *P. equorum* from a purportedly resistant isolate collected from foals on a farm in Ontario, Canada in which two consecutive treatments with ivermectin (11 weeks apart) failed to eliminate egg shedding. *Parascaris equorum* infected foals were blocked into cohorts of 2 based on FEC, and then allocated randomly within blocks to treated (ivermectin paste, 200µg/kg, $n = 6$) and untreated ($n = 5$) groups. FEC were performed from days -2 (before) to +13 days after treatment. During the entire period, no significant differences in *Parascaris* FEC were observed between treated and control groups. At the time of necropsy on day +13, strongylid FEC were uniformly zero in treated foals, while all untreated foals had positive strongylid FEC (arithmetic mean = 128.5 EPG). Ivermectin treatment resulted in 100%, 98.1% and 98.7% reductions of 2nd instars, 3rd instars, and total *Gasterophilus* specimens, respectively, but *P. equorum* worm counts were not significantly different between treated (137.5 ± 64.1) and untreated groups (153.6 ± 62.4). These data confirm that this isolate of *P. equorum* was resistant to ivermectin at the recommended, label dosage. It is advisable that efficacy of all anthelmintics be monitored in horses by performing FEC both before and after treatment.

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Cytokine responses to experimentally induced *Dirofilaria immitis* infections in cats. J.A. SPENCER[1]*, S. BARNEY[1], S. MAJOR[1], A.J. GUARINO[2], E.G. WELLES[1], C.M. JOHNSON[1], D.M. TILLSON[1], A.R. DILLON[1], B.L. BLAGBURN[1]. [1]COLLEGE OF VETERINARY MEDICINE, AND [2]COLLEGE OF EDUCATION, AUBURN UNIVERSITY, AUBURN, AL.

Cats are relatively resistant to establishment of heartworm infections. Studies have shown that the percentage of infective larvae developing to adult worms is low (0% to 25%) compared with dogs (40% to 90%). Adult heartworms in cats are smaller than those recovered from dogs, and have shorter life spans. This suggests that the immune response in cats is, at least in part, responsible for the developmental differences seen in feline heartworm infections. The major goal of this study was to examine selected cytokine immune responses to *D. immitis* infection in the cat. Cats were divided into 2 major groups: short term (8 mo) and long term (16 mo), and 3 subgroups. All cats in each group were infected with 100 L3 larvae. Group A (n=20) cats were treated monthly with selamectin. Group B (n=20) cats were treated every 2 weeks with ivermectin at 50 µg/kg beginning at 3 mo post-infection (p.i.) to mimic early death of L5 (sub-adult) worms. Group C (n=24) cats were untreated to mimic natural maturation of worms. Enriched circulating lymphocytes were collected at 4, 6 and 8 mo p.i. cDNA was prepared from these cells and assayed by reverse-transcriptase PCR for the presence of cytokine mRNA. Cytokines assayed for included G3PDH as housekeeping gene, γIFN, TNFα, IL-4, IL-5, IL-6 and IL-10. Results were expressed as presence or absence of the genes. There was a significant difference ($p<0.05$) seen in cytokine patterns in the ivermectin group immediately after treatment when compared with the selamectin and untreated groups. Gamma interferon and IL-5 expression were significantly decreased while IL-4 expression was significantly increased in the ivermectin group. Furthermore, IL-4 continued to be upregulated in this group over the course of the study, indicating an enhanced type 2 response to dead or dying worms.

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An examination of the published effects of preventative dosages of macrolide treatment on numbers of circulating microfilariae in dogs with patent heartworm (*Dirofilaria immitis*) infections. DWIGHT D. BOWMAN*[1], CHRISTOPHER J. TORRE[2]. [1]COLLEGE OF VETERINARY MEDICINE; AND [2]COLLEGE OF AGRICULTURE AND LIFE SCIENCES, CORNELL UNIVERSITY, ITHACA, NY.

Several studies have examined the effects of preventive heartworm treatments on the numbers of circulating microfilariae in the blood of dogs with patent heartworm infections. Ivermectin is known to have a microfilaricidal effect, and in three studies with the monthly treatment of microfilariae-positive dogs, five of six (Bowman *et al.*, 1992), seven of eight (Courtney *et al.*, 1998), and all five (McCall *et al.*, 1998) dogs became microfilariae-negative within 9 months or less after treatment was initiated. In this study the effects of ivermectin, milbemycin oxime, selamectin, and moxidectin (both the tablet and injectable formulations) will be compared as to the kinetics of clearance and the presence of microfilariae circulating in dogs after treatment (additional papers include: Lok *et al.*, 1989; Blagburn *et al.*, 1992; Hendrix *et al.*, 1992; Dzimianski *et al.*, 2002; Bowman *et al.*, 2002). This report will compare the data for those studies where dogs with patent infections have been treated with a macrolide at the preventive dose. An examination of these reports indicate that with these products, a majority of dogs remain microfilaremic for several months after the initiation of treatment and some for up to almost a year (and perhaps longer) after the beginning of preventative administration. In light of the fact that we have good alternatives to allowing the transmission of heartworms from microfilaremic dogs that are being given macrolides on a regular basis, it seems prudent to prevent their transmission. Prevention of transmission from such dogs can be done simply by insuring that veterinarians verify that dogs are heartworm negative before beginning preventive treatment and by treating infected dogs with melarsomine dihydrochloride before beginning macrolide clearance of microfilariae from the blood.

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Pathogenesis of feline heartworm infection. J. A. SPENCER[1]*, S. BARNEY[1], S. MAJOR[1], A.J. GUARINO[2], E. G. WELLES[1], C.M. JOHNSON[1], D.M. TILLSON[1], A.R. DILLON[1], B.L. BLAGBURN[1]. [1]COLLEGE OF VETERINARY MEDICINE, AND [2]COLLEGE OF EDUCATION, AUBURN UNIVERSITY, AUBURN, AL.

The arrival of immature *Dirofilaria immitis* L5 in the pulmonary vessels is associated with an intense eosinophilic pulmonary reaction. Clinical and radiographic signs may be present in this 3-6 month post-infection (p.i.) period. Because of the differences in host immune reactions and mortality of immature L5 worms, clinical signs and diagnosis of heartworm maybe different in the cat when compared with the dog. The goal of this study is to determine the response of the feline lung to the arrival and early death of young immature *D. immitis*. The majority of the clinical signs and life-threatening aspects of heartworm infection in cats are related to pulmonary disease. Cats were divided into 2 major groups: short term (8 mo) and long term (16 mo), and 3 subgroups. All cats were infected with 100 L3 larvae. Group A (n=20) cats were treated monthly with selamectin. Group B (n=20) cats were treated every 2 weeks with ivermectin at 50 μg/kg from 3 mo p.i. to mimic natural early death of L5 larvae. Group C (n=24) cats remained untreated to mimic natural maturation of worms. Cells obtained by bronchiolar lavage (BAL) were collected at 4, 6 and 8 mo p.i. and characterized by cell type and the presence of cytokine mRNA by reverse transcriptase PCR. Cytokines assayed for included G3PDH as housekeeping gene, γIFN, TNFα, IL-4, IL-5, IL-6 and IL-10. At 8 mo p.i. half of the cats from each group were euthanased and assessed for histopathological changes and presence of adult worms. Untreated cats showed BAL eosinophilia, and the most significant ($p<0.05$) decreases in TNFα, IL-5, IL-6, and IL-10

cytokine gene expression. The selamectin group showed the least changes and there were varying results in the ivermectin group. The following total numbers of worms were recovered from each group at necropsy: Group A: 0; Group B: 2; Group C: 45 plus 12 fragments. The most severe radiographic changes appeared to be in the ivermectin treated group. Histopathological results will be discussed in conjunction with BAL cytology and cytokine patterns.

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Discovery, structure, efficacy, and safety of dinotefuran, a third-generation neonicotinoid. A.H. AHN.* SUMMIT VETPHARM, FORT LEE, NJ.

The neonicotinoids are synthetic insecticides with a structure and mode of action similar to those of nicotine. Since the discovery of the neonicotinoids in the late 1970s, several highly effective commercial products have been developed. First-generation products include imidacloprid, acetamiprid, and nitenpyram. Second-generation products include clothianidin and thiamethoxam. The current presentation reviews the discovery and basic characteristics of dinotefuran, a novel third-generation neonicotinoid with efficacy similar to that of earlier products. This novel new compound has a tetrahydro-3-furylmethyl group instead of the heterocyclic aromatic ring that was previously thought to be necessary for insecticidal activity among neonicotinoid compounds. Synthesis of dinotefuran was based on chemical manipulation of acetylcholine, which acts on the same receptor as nicotine but does not have a pyridine ring. Dinotefuran has broad insecticidal activity but contains no halogens or benzene rings, making it extremely safe for people, animals, and the environment. The oral and dermal LD50 in rodents is ≥2000 mg/kg. Laboratory studies have also shown that dinotefuran is non-teratogenic, non-genotoxic, non-carcinogenic, and non-sensitizing to the skin. This combination of safety and efficacy makes dinotefuran an ideal candidate for control of ectoparasites of veterinary importance.

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Cat flea susceptibility To Imidacloprid: Results of A 5-year monitoring initiative. B. BLAGBURN[1]*, R. BOND[2], R. ARTHER[3], I. DENHOLM[4], M. DRYDEN[5], J. HOSTETLER[3], H. MEHLHORN[6], N. MENCKE[3], P. PAYNE[5], M. RUST[7], I. SCHROEDER[3], M. VAUGHN[3]. [1]AUBURN UNIVERSITY, [2]ROYAL VETERINARY COLLEGE, [3]BAYER HEALTHCARE ANIMAL HEALTH, [4]ROTHAMSTED RESEARCH, UK, [5]KANSAS STATE UNIVERSITY, [6]HEINRICH-HEINE UNIVERSITY, GERMANY, [7]UNIVERSITY OF CALIFORNIA, RIVERSIDE.

A flea larval bioassay was developed to monitor the susceptibility of fleas (*Ctenocephalides felis*) to imidacloprid (Advantage® Bayer HealthCare). The assay was validated using both laboratory and field isolates of *C. felis*. Flea eggs representing different field isolates of *C. felis* were collected by veterinarians in 21 states in the United States (US) and in the United Kingdom (UK) and Germany. A total of 972 flea isolates were obtained from during the 5-year study period. Of the 972 isolates received, 768 contained sufficient numbers of eggs to conduct the larval bioassay. Greater than 5% survival occurred for only 6 of the 768 field isolates that were evaluated. Further evaluation and analysis of these isolates demonstrated that they were not significantly different in their susceptibility to imidacloprid from the reference strains field isolates used to develop the assay. Also, at this point there do not appear to be any differences in susceptibility to imidacloprid of cat flea field isolates from the US, the UK and Germany. Collections of field flea isolates will continue in attempt to detect and document any change in the susceptibility of field flea populations to imidacloprid.

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Title and abstract not provided. DAVID DUFFY, UNIVERSITY OF HAWAII, HONOLULU, HI, U.S.A.

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Parasites in paradise - impacts of parasitic diseases on Hawaii's native species. THIERRY M. WORK*. US GEOLOGICAL SURVEY, NATIONAL WILDLIFE HEALTH CENTER, HONOLULU FIELD STATION, HAWAII, USA.

Hawaii has the largest *per capita* number of endangered animals and plants of any state. Loss of habitat and introduced predators have been the two factors most incriminated for declines of native species, however, diseases also play a significant role. Parasites are the most commonly encountered infectious diseases in Hawaiian wildlife. The classic and most publicized example is avian malaria that was introduced in Hawaii and that resulted in declines of native Hawaiian honeycreepers and reductions of their native range. Similarly, toxoplasmosis was probably partly responsible for the decline of endangered Hawaiian crow and for the recent removal of this bird from its native range. Toxoplasmosis continues to affect assorted native birds such as Hawaiian geese and seabirds albeit in low numbers. The nematode *Echinuria* sp. has been associated with significant mortalities of the critically endangered Laysan duck in the Northwestern Hawaiian islands and played a role in methods to repatriate this bird to its former native range. Adverse impacts of parasitic diseases extend beyond the shores to marine habitats. Sea turtles are commonly affected by 4 species of vascular flukes that can cause significant pathology, and occasionally, mortality. The blue lined snapper, introduced from the Marquesas into Hawaii in the 1950s has high prevalence of infection with *Goussia*, and this parasite is shared with native goatfish prompting the question of whether the parasite was introduced with snappers into Hawaii. Even invertebrates aren't spared. One of the most common diseases in Hawaiian corals is a trematode, and a ciliate probably contributed to a mass mortality of sea urchins in Johnston Atoll, Central Pacific.

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