

Proceedings of the

50th Annual Meeting of the Austrian Society of Tropical, Medicine, Parasitology and Migration Medicine

"50 years ÖGTPM - changes and challenges"



17th - 19th November 2016

Museum of Natural History, Vienna, Austria

www.ögtpm.at

Meeting Venue:

Naturhistorisches Museum Wien Natural History Museum Vienna

Burgring 7, 1010 Wien

Umschlagbild: Naturhistorisches Museum Wien/Natural History Museum Vienna Foto: © Kurt Kracher

Dear colleagues and friends,

The 50th Annual Meeting of the Austrian Society for Tropical Medicine, Parasitology and Migration Medicine is approaching and will be held - as in 2015 – at the Natural History Museum in Vienna from November 17-19, 2016.

As this year we celebrate a special anniversary, the motto of the conference is **"50 years ÖGTPM -**



changes and challenges". Since the first meeting of ÖGTPM in 1966 an incredible scientific progress in all fields covered by our society can be noted and we will try to reflect in part this progress in our meeting.

Again we were able to invite several distinguished speakers from various fields, among them Dennis Shanks from Australia who will give a special presentation on Malaria and Robert Steffen, the godfather of travel medicine, which has evolved to a special branch in medicine during the last four decades.

The scientific program will cover a diversity of topics, including parasitology, tropical medicine, microbiology and travel medicine and will give opportunity to researchers to present their last scientific results. The meeting is open for submission of presentations in any aspect of tropical medicine, parasitology and migration medicine. And we particularly encourage young researchers and scientists to submit their work: Again a poster price and a junior award for the best presentation will be sponsored.

As in the preceding years submissions may be either presented as oral presentations or in a chaired poster session giving authors the opportunity to present their work.

On this occasion I would like to thank our sponsors, who are again supporting our meeting and without their funding this meeting would not be possible.

Looking forward to seeing you in Vienna for an exciting anniversary meeting

Best regards

Herwig Kollaritsch (president of ÖGTPM)

PROGRAMME

Applications who can be voted for Junior Award/Poster Prize a marked with an "*" in the program.

THURSDAY, NOVEMBER 17th

08:30 – 09:00 Registration

09:00 - 09:10 Welcome and Introduction

09:10 - 12:40 VECTOR-BORNE DISEASES

(chaired by: J. Walochnik, G. Duscher)

09:10 – 09:55 KEYNOTE LECTURE em.O.Univ.Prof.Dr. Franz Xaver Heinz "Viruses and their importance for mankind – focus on VBD"

9:55 – 10:10 Franz Allerberger: "Austrian communicable diseases prepardeness plan"

10:10 – 10:25 Gerold Stanek: "Lyme borreliosis: conflict points in diagnosis and treatment"

10:25 - 10:40 Martin Weiler: "Tick screening on military training sites in Austria"

10:40 – 11:25 Coffee break

11:25 – 11:40 Erich Tauber: "Development of a chikungunya vaccine using a recombinant measles virus vaccine vector"

11:40 – 11:55 Aleksandar Potkonjak: "Molecular detection of flea-borne pathogens in *Ctenocephalides felis* collected from cats in Serbia: Preliminary results"

*11:55 – 12:10 Tomáš Macháček: "Nitric oxide and proinflammatory cytokines are produced by glial cells exposed to *Trichobilharzia regenti*, a neuropathogenic schistosome causing cercarial dermatitis"

12:10 – 12:25 Jan Pankrác: "Effect of radicals and other reactive species on *Trichobilharzia regenti*, a neuropathogenic avian schistosome"

12:25 - 12:40 Convenience break

12:40 – 13:40 LUNCH SYMPOSIUM "UNTERSCHÄTZTE PARASITEN" (in German): (Organized by INSTAND & ÖQUASTA)

(Vorsitz: K. Janitschke, H. Auer)

Ingrid Reiter-Owona: "Trichomonas beim Mann" Uwe Gross: "Beeinträchtigen Toxoplasmen unser Verhalten" Julia Walochnik: "Giardia in Mitteleuropa"

13:40 – 14:00 Convenience break

14:00 - 15:00 MULTI-RESISTANT BACTERIA

(chaired by: M. Haditsch, B. Haas)

14:00 – 14:45 KEYNOTE LECTURE Univ. Prof. Dr. Ivo Steinmetz (Institute for Hygiene, Microbiology and Environmental Medicine, Graz) "Melioidosis: a globally neglected tropical disease"

14:45 – 15:10 Alexander Indra: "Multiresitant tuberculosis - European importance"

15:10 - 16:00 Coffee break

16:00 - 18:15 MIXED TALKS

(chaired by: M. Duchêne, R. Moser)

16:00 – 16:30 Mzia Turashvili: "Health of humanitarian workers"

*16:30 – 16:45 Aruna Shrestha: "Cloning, expression and molecular characterization of a *Cystoisospora suis* specific uncharacterized merozoite protein"

*16:45 – 17:00 Mirjana Drinić: "Immunomodulatory properties of *Toxoplasma gondii* tachyzoites extract are potentially linked to carbohydrate components and lead to suppression of allergic airway inflammation in mice"

*17:00- 17:15 Adnan Hodžić: "Cardiorespiratory parasites in red foxes (*Vulpes vulpes*) from Bosnia and Herzegovina"

17:15-17:30 Alisa Gruden-Movsesijan: "Immunology lesson given by "Old Friend" *Trichinella spiralis*"

17:30 – 17:45 Walter Glawischnig: "OIE Twinning – a valuable concept to control *Trichinella* infections in animals and foodstuff in Tanzania"

17:45 – 18:00 S. Steuber: "Prevalence of insecticide resistance house flies (*Musca domestica*) on pig farms in Schleswig-Holstein, Germany"

18:00 – 18:15 Steffen Rehbein: "Parasites of the Alpine Ibex (*Capra i. ibex*) in North Tyrol – faunal diversity and abundance"

18:30 – 20:30 GET TOGETHER (NHM) "Dinosaurier Saal"

FRIDAY, NOVEMBER 18th

08:30-09:00 Registration

09:00 – 11:45 "YOUNG PARASITOLOGISTS SESSION" : VECTORS AND VECTOR BORNE DISEASES IN EUROPE"

(chaired by: H.P.Fuehrer, U. Fürnkranz)

9:00 – 9:45 KEYNOTE LECTURE Ana Margarida Alho: "Dirofilariosis, an overview and a multidisciplinary approach conducted in an European country"

*09:45 – 10:00 Viktoria Wimmer: "Filarioid helminths in mosquitoes from the Danube Delta/Romania and the analysis of these vectors for potential vector competence"

*10:00 – 10:15 Sarah Übleis: "Molecular epidemiology of mosquito-borne diseases in Eastern Austria"

*10:15 – 10:30 Carina Zittra: "Spatiotemporal mosquito species distribution in Eastern Austria driven by environmental parameters - final results"

10.30 - 11:15 Coffee break

*11:15 – 11:30 David Ebmer: "Morphological and molecular identification of *Synhimantus (Synhimantus) laticeps* (Rudolphi, 1819) Railliet, Henry et Sisoff, 1912 (Nematoda, Acuariidae) from the barn owl (*Tyto alba*) and the common kestrel (*Falco tinnunculus*) in Austria"

11:30 – 11:45 Aline Lamien-Meda: "Novel approaches to diagnosing malaria. Development and validation of a High Resolution Melting Curve Analysis assay for human malaria diagnosis"

*11:45 – 12:00 Susanne Reier: "Molecular genetics and morphological identification of Austrian Acanthocephala (Kohlreuther, 1771)"

12:00 – 13:00 LUNCH SYMPOSIUM II "SEX und PARASITEN " (in German): (Organized by INSTAND & ÖQUASTA)

(Vorsitz: W. Graninger, H. Aspöck)

Heimo Lagler: "Sexuell übertragbare Viruserkrankungen- HepB/C HIV, HPV,Zika-Update 2016"

Anton Aebischer: "Neu überarbeiteter Ratgeber für Ärzte zu Skabies" Herbert Auer: "Filzläuse (pubic lice, scrab lice)"

13:00 - 15:00 MALARIA/MILITARY MEDICINE

(chaired by: H. Nödl, G. Rosenmayr)

13:00 – 13:45 KEYNOTE LECTURE Prof. Dennis Shanks (Australian Army Malaria Institute): "The military and malaria in the last century: epidemics, deployments and drug development"

13:45 - 14:15 Harald Nödl: "Malaria - Past, Present and Future"

14:15 – 14:30 Erich Schmutzhard: "Adjunctive therapies in life-threatening *P. falciparum* malaria"

*14:30 – 14:45 Elisa Reiterer: "Children with hearing loss secondary to severe and cerebral malaria do not regain full cochlear function after four years – a follow-up study."

14:45 – 15:00 Rupert Bliem: "EBOLA: A personal experience report based on an EMLab deployment to western Africa"

15:00 - 15:30 Coffee break

15:30 – 17:45 POSTER SESSIONS (NHM poster walls, near Cafeteria) (chaired by: I. Schabussova, M. Drinić)

P1 Ursula Fürnkranz et al.: In co-cultures of Mycoplasma hominis with Trichomonas vaginalis type strain G3, displacement of M. hominis strain Mh475 by MhAKH led to loss of mycoplasmal TET-resistance and loss of parasitic stress response

***P2** Christian Gatterer et al.: Door-to-door survey: Epilepsy in a rural area in southern Tanzania. Prevalence, semiology and treatment gap of epilepsy in Mahenge, Ulanga

P3 Barbara Hinney et al.: Comparison of methods for the quantitative analysis of Strongyle eggs in horse faeces

***P4** Teresa Pfeiffenberger et al.: Nodding Syndrome in Mahenge, Tanzania, a 10year follow up.

P5 Trattner Elisabeth et al.: Multiple Intestinal Protozoal Parasites and Filariasis in a Migrant from Bangladesh

***P6** Anna-Margarita Schötta et al.:Comparison of tick-screening methods for pathogen detection and impact on results

17:45 EVENING EVENT in Presentation Hall "Kinosaal"

In memoriam Prof. Walther Wernsdorfer (held by H. Nödl)

Music: Johann Sebastian Bach Air aus der Orchestersuite Nr. 3 in D-Dur, BWV 1068

> Nima Naraghi, 1. Violine Isabelle Reinisch, 2. Violine Georg Schröfl, Viola Christoph Aspöck, Violoncello

Horst Aspöck: "50 Jahre Österreichische Gesellschaft für Tropenmedizin / und Parasitologie / und Migrationsmedizin – in Bildern und Anekdoten "

Music: Antonín Dvořák Streichquartett in F-Dur, op. 96, das "Amerikanische"

> Nima Naraghi, 1. Violine Isabelle Reinisch, 2. Violine Georg Schröfl, Viola Christoph Aspöck, Violoncello

Junior Award

Poster prizes

19.30 DINNER (NHM, Upper Hall)

SATURDAY, NOVEMBER 19th

FORTBILDUNG ÄRZTE/APOTHEKER (in German language)

08:30 - 09:00 Registrierung

9:00 - 10:30 REISEMEDIZIN I

(Vorsitz: U. Wiedermann, H. Kollaritsch)

09:00 – 09:45 KEYNOTE LECTURE: Prof.Robert Steffen, WHO, Univ.Zürich) "Reisemedizin: Aufgaben und Herausforderungen im Wandel der Zeit"

09:45 - 10:05 Ursula Wiedermann:"Impfungen in der Reisemedizin - update"

10:05 - 10:30 Herwig Kollaritsch: "update Weltseuchenlage"

10:30 – 11:00 Kaffeepause

11:00 - 12:15 REISEMEDIZIN II

11:00 -11:30 Horst Aspöck: "Arthropoden: Fakten und Mythen. Herausforderungen der Medizinischen Entomologie im 21. Jahrhundert"

11:30 – 12:15 Herwig Kollaritsch und Martin Haditsch: Reisemedizinisches Quiz mit Televoting

12:30 Ende

Abstracts in chronological order:

Viruses and their importance for mankind – focus on vector-borne diseases

Franz X. Heinz

Department of Virology, Medical University of Vienna, 1090 Vienna, Austria

franz.x.heinz@meduniwien.ac.at

Viruses have caused some of the most devastating epidemics in human history, with smallpox and Spanish flu being prominent examples of the enormous death tolls that can result from virus infections. Understandably, viruses attract our attention primarily because of their capacity to induce disease but at least some of them were also major driving forces of biological evolution. One of the most spectacular examples in this context is the acquisition of retroviral proteins (syncytins) that can mediate the fusion of cellular membranes. These virus-derived genes allowed the formation of a specific multinucleated cellular structure, the syncytiotrophoblast, a key tissue of placentas and essential for the evolution of mammals.

New human viruses can emerge through trans-species transmissions at the animal-human interface. SARS, MERS, Ebola, Influenza, and most importantly HIV are prominent examples for the threats imposed by newly emerging viruses. Virus jumps from one species to another, however, are severely impeded by the dependency of the virus on cellular molecules for replication (the interactome) and the presence of cellular restriction factors that provide a barrier of resistance to new viruses. I will use the example of HIV to demonstrate the arms race between host and virus that led to the generation of a new human-to-human transmissible virus originating in apes.

Arthropod-borne (ARBO) viruses are a special case because they require efficient replication in phylogenetically very distant animal species, invertebrates and vertebrates. Some ARBO viruses can cause explosive epidemics with millions of people infected annually. Dengue and Chikungunya are prominent examples. Recently, the previously largely cryptic Zika virus was added to this list and caused completely new threats to global health because of several unanticipated properties. It caused massive outbreaks in the Americas which before 2014 were completely free of this virus. Although most of the infections result in either no or only mild clinical symptoms, congenital infections and severe malformations were causally linked to Zika virus, a property not previously described for any ARBO virus. The same also holds true for sexual transmission which is unique to Zika virus and poses problems to people living in or returning from Zika-endemic areas. Furthermore, discomforting animal experiments point to possible adverse effects on testicles and male fertility. Different kinds of vaccines are being developed and first clinical trials in humans are in progress.

Austrian communicable diseases prepardeness plan

Franz Allerberger

Austrian Agency for Health and Food Safety (AGES), Vienna, Austria

franz.allerberger@ages.at

The Austrian Agency for Health and Food Safety (AGES) is a company of the Republic of Austria, owned by the Austrian Federal Ministry of Health and Women's Affairs and the Federal Ministry of Agriculture, Forestry, Environment and Water Management. AGES supports the management of the federal ministries in questions relating to public health, animal health, food safety, medical and drug safety, and food security by providing professional and independent scientific expertise. The principal legal basis for addressing the risks of the spread of infectious diseases are the Epidemics Act ("Epidemiegesetz 1950") and the Zoonoses Act 2005 ("Bundesgesetz zur Überwachung von Zoonosen und Zoonoseerregern (Zoonosengesetz)"). Since January 2014 Austrian laboratories have a legal obligation to report occurrence of communicable diseases electronically into the so-called "Epidemiologische Meldesystem (EMS)", centrally operated by AGES. Attended physicians and hospitals have the choice to report either electronically, by fax or by surface mail. Risk management of health authorities depends heavily on efficient surveillance, alert, and response activities, which aim to support the local districts ("Bezirksverwaltungsbehörden") formally in charge of providing public health responses. In addition to the provisions dealing with the detection and response to specific events (e.g. small pox preparedness plan; measles standard operating procedure), the Austrian communicable diseases preparedness plan also utilizes situation-based surveillance systems related to recognize the spread of diseases, e.g. the syndromic surveillance implemented in migrant reception centres in Austria in 2015.

Lyme borreliosis: conflict points in diagnosis and treatment

Gerold Stanek

Institute for Hygiene and Applied Immunology, Medical University of Vienna, Vienna, Austria;

gerold.stanek@meduniwien.ac.at

Lyme borreliosis is the most frequent tick-borne disease in the northern hemisphere. The prevalent pathogens in Europe are the genospecies *Borrelia afzelii, B. garinii, B. bavariensis,* and *B. burgdorferi* sensu stricto. Reservoir hosts usually are small mammals and birds; vectors are ticks of the genus *Ixodes: I. ricinus* and *I. persulcatus* in Europe, *I. persulcatus* in Asia, and *I. scapularis* and *I. pacificus* in North America. The disease manifests most frequently at the tick bite site as the skin infection erythema migrans. Disseminated infections comprise the nervous system, joints and heart and only very rarely other organs. Acrodermatitis chronica atrophicans, arthritis, and persistent infections.

Diagnostic support by the laboratory is primarily based on serology. For the time being two-tier testing is recommended and detection of intrathecally produced antibodies in cases of suspected neuroborreliosis. Antibiotic treatment is suggested for 10 days to two weeks in localised and disseminated disease, and for a maximum of four weeks in chronic disease. Generally, the prognosis for a favourable outcome is excellent.

However, high sero-prevalence, usage of serology without clinical suspect, persistent IgM antibodies, usage of serology to control the therapeutic outcome result in arbitrary interpretations of serological results. Moreover, not sufficiently validated cellular tests, CD57 cell count, and dark field or phase contrast microscopy are offered.

Finally, there is another world of "Lyme" which imparts another picture of the disease and treatment schedules of extraordinary length.

Tick screening on military training sites in Austria

Martin Weiler^{1, 3}, Georg Duscher², Peter Hufnagl⁴, Julia Walochnik¹

¹ Institute of Specific Prophylaxis und Tropical Medicine, Medical University of Vienna, Vienna, Austria

² Institute of Parasitology, University of Veterinary Medicine, Vienna, Austria

³ CBRN-Defense School, Austrian Armed Forces, Korneuburg, Austria

⁴Austrian Agency for Health and Food Safety (AGES), Vienna, Austria

martin.weiler@bmlv.gv.at

Ticks are the most relevant vectors of pathogens in Austria. As military training is conducted primarily in the field, tick exposure cannot be prevented entirely. For this reason an evidence-based medical risk management is crucial for preventive measures. The goal of this study was to collect baseline data for a follow-on risk assessment for various training activities in different seasons at the training sites. Ticks were collected from all 54 active training sites in Austria. On two locations ticks were sampled on a weekly basis to track the tick activity during the year. This revealed the expected peaks in spring and autumn, but also the effects of flooding for one season. To date, more than 7000 ticks have been collected. Ticks were found at all training sites, even at an altitude of 1,450 m a.s.l. *Ixodes ricinus* was the predominant species (92%), followed by *Haemaphysalis concinna* (7%) and *Dermacentor reticulatus* (<1%). The spectrum of pathogens carried by the

collected ticks was evaluated by PCR. The following pathogens are included in the screening: TBE-virus, *Borrelia burgdorferi, Brucella* spp., *Coxiella burnetii, Francisella tularensis,* and *Babesia* spp. Until now, screenings (pools of 10 *Ixodes ricinus* nymphs each) were negative for *Francisella tularensis* and *Coxiella burnetii.* TBE-virus was found in 0,01% (2 out of 150) of the tested pools. 66% of the pools were positive for *Borrelia burgdorferi*.

Development of a chikungunya vaccine using a recombinant measles virus vaccine vector

Erich Tauber^{*}, Katrin Ramsauer, Matthias Müllner

Themis Bioscience Gmbh, Muthgasse 11/2, 1190 Vienna, Austria

*erich.tauber@themisbio.com

Recurrent outbreaks of chikungunya virus suggest the development of a vaccine against this emerging disease. We have developed a vaccine by inserting nucleotide sequences encoding the Chikungunya virus capsid and envelope structural proteins into a measles vaccine strain virus to produce the candidate vaccine MV-CHIK.

To evaluate the optimal dose of MV-CHIK in regard to immunogenicity, safety and tolerability we performed an observer blinded, block-randomized, active and placebo-controlled, dose escalation, phase 1 trial in 42 healthy volunteer subjects. A series of increasing doses of MV-CHIK (1.5x104 - 3x105 TCID50 per individual) was injected intramuscularly in three groups of volunteers. The commercially available measles containing virus vaccine Priorix® was used as active control. Subjects within a cohort (MV-CHIK: n=12, Priorix: n=2/ Cohort) were block-randomized to receive a booster immunization either on day 28 or day 90 after the first vaccination. The primary endpoint was the presence of neutralizing antibodies on day 28 assessed by 50% plaque reduction neutralization titer (PRNT50) assay. Secondary endpoints included anti-vector immunity (Chikungunya vs. measles).

36 participants were included into the per-protocol population. MV-CHIK raised neutralizing antibodies in all dose cohorts after a single immunization with seroconversion rates of 44% (4/9) low dose, 92% (11/12) medium dose and 90% (9/10) high dose. The MV-CHIK immunogenicity was not affected by pre-existing anti-measles immunity. The second vaccination resulted in a 100% seroconversion across all MV-CHIK cohorts with an overall good safety profile. No vaccination-related serious adverse events occurred.

MV-CHIK showed good immunogenicity, even in the presence of anti-vector immunity, was safe and had an overall acceptable tolerability profile. MV-CHIK is the first promising MV-based candidate vaccine entering humans. The promising vaccine candidate is currently being tested in phase 2 clinical trials to show tolerability, safety and immunogenicity in the presence of anti-vector immunity in a larger population with the goal to determine a dose and schedule for Phase 3 clinical development.

References

Ramsauer K, Schwameis M, Firbas C, Müllner M, Putnak RJ, Thomas SJ, Despres P, Tauber E, Jilma, B Tangy F. Lancet Infectious Diseases. 2015;

Molecular detection of flea-borne pathogens in *Ctenocephalides felis* collected from cats in Serbia: Preliminary results

<u>Aleksandar Potkonjak</u>^{1*}, Adnan Hodžić², Vuk Vračar¹, Dragan Bacić³, Aleksandra Petrović⁴, Aleksandra Ignjatović Ćupina⁴, Vesna Lalosević¹, Joachim Spergser⁵, Georg G. Duscher²

¹ Department of Veterinary Medicine, Faculty of Agriculture, University of Novi Sad, Trg Dositeja Obradovića 8, 21000 Novi Sad, Serbia

² Department of Pathobiology, Institut of Parasitology, Universiy of Veterinary Medicine Vienna, Veterinaerplatz 1, 1210 Vienna, Austria

³ Department of Infectious Animals Diseases and Diseases of Bees, Faculty of Veterinary Medicine, University of Belgrade, Bulevar oslobođenja 18, 11000 Belgrade, Serbia

⁴ Department of Environmental and Plant Protection, Faculty of Agriculture, University of Novi Sad, Trg Dositeja Obradovića 8, 21000 Novi Sad, Serbia

⁵ Department of Pathobiology, Institute of Microbiology, University of Veterinary Medicine Vienna, Veterinaerplatz 1, 1210, Vienna, Austria

*ale@polj.uns.ac.rs

Flea-borne infections are considering emerging or re-emerging, and their prevalence has been increased worldwide. The aim of the present study was to determine the occurrence and distribution of *Bartonella* spp., *Rickettsia* spp. and haemotropic mycoplasmas in fleas collected from companion and stray cats in five localities (Belgrade, Novi Sad, Kać, Bačka Palanka and Despotovo) in Serbia. A total of 519 cat fleas (*Ctenocephalides felis*) collected were merged in 113 pools, and 37 (32.7%), 10 (8.8%) and 14 (12.3%) of the pools were found to be PCR positive for *Bartonella* spp., *Rickettsia* spp. and haemotropic mycoplasmas, respectively. Fleas from all five localities were infected with pathogens, and minimal infection rate (95% confidence interval) for *Bartonella* spp. was 7.13% (4.9–9.3%); for *Rickettsia* spp. 1.9% (0.7–3.1%); and for haemotropic mycoplasmas it was 2.7% (1.3–4.0%). Results show that these pathogens circulating in cats fleas in Serbia. Further researches are needed to identify actual prevalence of flea-borne pathogens in cat fleas.

References

1. Bitam I, Dittmar K, Parola P, Whiting MF, Raoult D. Fleas and flea-borne diseases. Int J Infect Dis. 2010;14(8):667-76.

2. Barrs VR, Beatty JA, Wilson BJ, Évans N, Gowan R, Baral RM, Lingard AE, Perkovic G, Hawley JR, Lappin MR. Prevalence of *Bartonella* species, *Rickettsia felis*, haemoplasmas and the Ehrlichia group in the blood of cats and fleas in eastern Australia. Aust Vet J, 2010;88(5):160-5.

Nitric oxide and proinflammatory cytokines are produced by glial cells exposed to *Trichobilharzia regenti*, a neuropathogenic schistosome causing cercarial dermatitis

Tomáš Macháček^{*}, Lucie Panská, Petr Horák

Department of Parasitology, Charles University, Prague, 12843, Czech Republic

*tomas.machacek@natur.cuni.cz

Immunological processes ongoing during helminth-caused neuroinfections and their role in tissue pathology and parasite elimination are usually poorly recognized. We examined the role of nitric oxide (NO) and several cytokines produced by astrocytes and microglia, the immunocompetent cells of the CNS, after exposure to *Trichobilharzia regenti*. It is a neurotropic schistosome of birds in which it migrates *via* the CNS. This is accompanied by neuromotor disorders, and inflammation of the affected nervous tissue is triggered. Moreover, activation of astrocytes and microglia was also noted around the schistosomula migrating to the CNS in mice in which the parasites die, because mice represent an accidental (incompatible) host.

Consequently, we *in vitro* examined the capability of astrocytes and microglia to promote or regulate tissue inflammation, and to kill the parasites. In particular, we measured production of NO and both proinflammatory (IL-6, TNF- α) and antiinflammatory (IL-10, TGF- β 1) cytokines by murine astrocyte and microglia primary cultures 48 hours after exposure to selected stimuli of *T. regenti* origin. These were living *T. regenti* schistosomula-like stages prepared *in vitro* (LS), a soluble fraction of their homogenate (HSF) and recombinant cathepsins B1.1 and B2 of *T. regenti* (CB1.1 and CB2) which are prevailing proteases expressed by migrating schistosomula.

NO production was triggered by HSF, CB1.1 and CB2 in astrocyte cultures, and also by HSF in microglia cultures. As NO was not produced in any culture after exposure to LS, it is probably not responsible for parasite killing but rather may contribute to neural damage. *T. regenti*-derived stimulants also increased production of proinflammatory cytokines IL-6 and TNF- α in astrocyte and microglia cultures, but no significant changes were noticed in production of anti-inflammatory cytokines IL-10 and TGF- β 1. This suggests an important role of these cells in maintaining the tissue inflammation during the infection.

Acknowledgements: Grant Agency of Charles University (GAUK 250198), Czech Science Foundation (GAČR 13-29577S), institutional/departmental support (UNCE 204017, PRVOUK P41, SVV 260319/2016).

Effect of radicals and other reactive species on *Trichobilharzia regenti*, a neuropathogenic avian schistosome

Jan Pankrác^{1*}, Martin Kašný^{1,2}, Petr Horák¹

¹ Department of Parasitology, Faculty of Science, Charles University, Prague 128 43, Czech Republic

² Department of Botany and Zoology, Faculty of Science, Masaryk University, Brno 625 00, Czech Republic

*pankra@seznam.cz

Previous research showed that trematodes can regenerate certain body structures (e.g., reproductive organs, tegument, and muscles) which have been damaged mechanically, by the action of praziquantel, or due to cultivation under suboptimal conditions. However, there is still limited knowledge of trematode response to tissue damage caused by highly reactive oxygen molecules produced by host immune cells. In our research, we aimed at pathological changes induced by exposure of *T. regenti* schistosomula to hydrogen peroxide (H_2O_2), and consequent response of the parasite.

Schistosomula of *T. regenti* were exposed to 1.6 mmol/l solution of H_2O_2 , and cultured in schistosome culture medium (SCM) 169. Protein and DNA oxidative damages were localized immunohistochemically by antibodies to 8-hydroxy-2'-deoxyguanosine and dinitrophenol. Fluorescent staining was performed on whole parasites 3 hours post-exposure to H_2O_2 . One day post-exposure, morphological changes in the surviving schistosomula were evaluated in histological sections stained with hematoxyline-eosin, and by electron microscopy. Changes in the amount of glycogen in tissues were determined by use of periodic acid-Shiff (PAS) reagent.

The mortality rate of 25-60 % per 24 hours was induced by exposure to 1.6 mmol/l H_2O_2 in SCM 169 for 15 minutes. Temporary immobilization and occasional shrinkage of schistosomulum body were observed during the exposure. Oxidative damage was detected in proteins of muscle fibers and tegument 3 hours post exposure, DNA damage was not proved. In schistosomula surviving one day post-exposure, PAS reagent and hematoxylin-eosin staining did not show clearly visible pathological changes. No ultrastructural changes were detected on the outer tegumental membrane (scanning electron microscopy), in tegument, mitochondria, and muscles (transmission electron microscopy). Absence of pronounced tissue damage in surviving schistosomula suggests that pathological changes resulting from exposure to reactive oxygen species are usually fatal, and the role of regeneration is limited.

Acknowledgements: Grant Agency of Charles University (GAUK 250198).

Trichomonas vaginalis: an underestimated sexual transmitted infection in men?

Ingrid Reiter-Owona¹*, Carolynne Schwarze-Zander², Thorsten Hornung³, Marijo Parcina¹, Achim Hoerauf¹

¹ Institute of Medical Microbiology, Immunology and Parasitology

² Department of Internal Medicine 1, University of Bonn, Germany

³ Department of Dermatology and Allergology

University Clinic of Bonn, 53105 Bonn, Germany

* reiter-owona@microbiology-bonn.de

In Germany, sexually transmitted infection (STI) rates are increasing during the last years. Increasing rates of syphilis are recorded predominately in men who have sex with men (MSM), increasing rates of *Chlamydia trachomatis* (CT) und *Neisseria gonorrhoeae* (GC) in the heterosexual population [1]. Information on the prevalence of *Trichomonas vaginalis*, worldwide considered as leading non-viral sexually transmitted infection, is sparse, especially in men. Infected men are often asymptomatic and considered as "reservoir" for female sexual partners. Current diagnostic methods of *T. vaginalis* in men rely on the in vitro cultivation of the parasite from urine specimens. Other test systems like antigen detection or molecular biological assays (PCRs) are often established for female specimens, only. We validated the BD MaxTM CT/GC/TV assay (real-time PCR) for the detection of *T. vaginalis* DNA in male urine. Subsequently, we applied the assay in urine samples of men attending different STI ambulances at the university clinics of Bonn.

Altogether, 197 urine samples (\geq 10 ml) from men aged 20-74 (mean 37) years were screened by PCR. 94 samples were collected from male heterosexuals (Group I) and 103 samples from MSM, gay or bisexual (Group II), with history of past HIV infection. Samples from Group I were also submitted to in vitro cultivation (OxoidTM Trichomonas Medium, min. 7 days). In none of the urine samples *T. vaginalis* was detected by PCR or culture. The BD MaxTM CT/GC/TV assay detected CT in 5 samples (5.3%) and GC in 4 samples (4.3%) out of Group I, and CT in 5 samples (4.9%) out of Group II.

The results indicate that *T. vaginalis* is not an underestimated infection in men. The results are in accordance with a low prevalence of *T. vaginalis* in female sex workers (3%) reported recently [2].

[1] http://www.geschlechtskrankheiten.de/aktuelle-informationen [2]https://www.rki.de/DE/Content/InfAZ/S/STI/Studien/KABPsurvSTI/KABPsurvSTI _inhalt.html

Können Toxoplasmen unser Verhalten beeinträchtigen?

<u>Uwe Groß^{1,*}</u>, Tobias Sommer-Blöchl², Christian Gaser³

¹Deutsches Konsiliarlabor für Toxoplasma, Institut für Medizinische Mikrobiologie, Universitätsmedizin Göttingen, Göttingen, Germany, D37075 ²Institut für Systemische Neurowissenschaften, Universitätsklinikum Hamburg-Eppendorf, Hamburg, Germany, D20248 ³Klinik für Neurologie, Universitätsklinikum Jena, Jena, Germany, D07747

*ugross@gwdg.de

Nicht zuletzt aufgrund seiner sehr geringen Spezifität für verschiedene Zwischenwirte (z. B. Nagetiere, Vögel, Schlachttiere, aber auch Menschen) gehört Toxoplasma gondii zu den erfolgreichsten Parasiten unserer Erde. Um eine genetische Variabilität aufrecht zu erhalten, ist der Parasit darauf angewiesen, sich geschlechtlich im Darm der als Endwirte fungierenden Katzen (Felidae) zu vermehren. Doch warum sollte sich eine infizierte Maus freiwillig fressen lassen? Zahlreiche Versuchsanordnungen haben mittlerweile eindeutig belegt, dass Toxoplasmen das Verhalten infizierter Mäuse manipulieren und hierfür tief in die Trickkiste greifen. So befallen die Parasiten in Mäusen und Ratten vor allem die Mandelkernregion. In dieser zum limbischen System zählenden Hirnregion werden u.a. Fluchtreflexe verarbeitet. Durch diese Lokalisation und einen Eingriff in die Kommunikation manipulieren Toxoplasmen das Verhalten interneuronale infizierter Nagetiere: Der Geruch von Katzenurin löst nicht mehr einen Fluchtreflex aus, sondern führt sogar dazu, dass Toxoplasma-infizierte Mäuse offensichtlich angelockt und so zur leichten Beute werden [1]. Doch warum infizieren Toxoplasmen auch beim Menschen vor allem das Gehirn? Die Antwort mögen die Ergebnisse von Versuchen an Schimpansen bringen: Auch hier zeigte sich, dass Toxoplasma-infizierte Schimpansen im Vergleich zu nicht infizierten Kontrolltieren ein signifikant verstärktes Interesse für Leopardenurin zeigten [2]. Bedenkt man, dass die entwicklungsgeschichtliche Trennung der Vorfahren des modernen Menschen und von Schimpansen gerade einmal vor etwa 8 Millionen Jahren lag, ist zu erwarten, dass Toxoplasmen auch das Verhalten des Menschen beeinträchtigen. Und in der Tat mehren sich die Hinweise, dass Toxoplasmen unser Verhalten zu manipulieren vermögen [3]. Aktuell untersuchen wir, ob und ggf. welche neurobiologischen Faktoren bei Toxoplasma-infizierten Menschen im Vergleich zu nicht-infizierten Kontrollen Auffälligkeiten zeigen.

References

1. Vyas A, Kim, S-K, Giacomini, Boothroyd JC, Sapolsky R. Behavioral changes induced by *Toxoplasma infection* of rodents are highly specific to aversion of cat odors. Proc Natl Acad Sci 2007; 104:6442-6447.

2. Poirotte C, Kappeler PM, Ngoubangoye B, Bourgeois S, Moussodji M, Charpentier MJ. Morbid attraction to leopard urine in *Toxoplasma*-infected chimpanzees. Curr Biol 2016; 26:R98-99.

3. Lindová J, Novotná M, Havlícek J, Jozífková E, Skallová A, Kolbeková P, Hodný Z, Kodym P, Flegr J. Gender differences in behavioural changes induced by latent toxoplasmosis. Int J Parasitol 2006; 36:1485-1492.

Giardia in Central Europe

Julia Walochnik

Institute for Specific Prophylaxis and Tropical Medicine, Medical University of Vienna, Vienna, Austria;

julia.walochnik@meduniwien.ac.at

Giardia spp. are flagellated protozoan parasites causing intestinal infections in a wide range of mammals, reptiles and birds. In Central Europe, approximately 2-4% of the population are infected, in several developing countries the prevalence can reach more than 40% whereby children up to 10 years of age are most affected. The life cycle involves a fragile, feeding and dividing trophozoite and a highly resistant cyst which can survive harsh environmental conditions for long periods of time. The infection is acquired orally by ingestion of the cysts with contaminated water or food or by direct fecal-oral transmission. Excystation results in the release of motile trophozoites which multiply in the small intestine causing asymptomatic or mild self-limiting infections in most cases or acute to severe gastroenteritis, occasionally resulting in malabsorption and long term growth retardation in children. The cysts, in acute cases the trophozoites, can be detected in fresh stool specimens, usually even without concentration or staining. Generally, the investigation of consecutive samples is recommended. To increase sensitivity, stool samples can also be investigated for Giardia antigens or DNA. Giardiosis is often under-reported due to the self-limiting nature of infections.

G. duodenalis (synonyms *G. intestinalis* and *G. lamblia*) consists of eight genetic assemblages (A-H), which today are regarded as separate species each with varying host specificities. Assemblage A, corresponding to *G. duodenalis*, and assemblage B, corresponding to *G. enterica*, are responsible for the majority of human infections. Both, assemblages A and B, are zoonotic and infect a wide range of animals including dogs, cats, livestock and wildlife. In Austria, a high level of genetic diversity of strains isolated form human patients has been detected, whereby assemblage B was found most frequently. This suggests that the majority of human infections are imported and that endemic anthroponotic transmission plays a minor role in Austria.

Melioidosis: a globally neglected tropical disease

Ivo Steinmetz

Institut für Hygiene, Mikrobiologie und Umweltmedizin, Medizinische Universität

Graz

ivo.steinmetz@medunigraz.at

The gram-negative bacterium Burkholderia pseudomallei can be found in the environment of the tropics and subtropics and causes the infectious disease melioidosis in humans and animals. Infection is acquired through inoculation, aerosols or ingestion. Although described more than a century ago, the precise worldwide distribution of Burkholderia pseudomallei is still unknown. The disease is known to be highly endemic in Southeast Asia and northern Australia. However, an increasing number of melioidosis case reports or environmental isolation of B. pseudomallei from other parts of Asia, Africa, the Caribbean, and Central and South America suggest a worldwide, but grossly underreported distribution of this pathogen. A recent study predicted an alarming number of 165,000 cases of human melioidosis per year worldwide, from which 89,000 people die. Melioidosis causes a wide range of acute or chronic clinical manifestations, including pneumonia, abscesses in various organs, neurological manifestations, or severe septicemia. B. pseudomallei grows intracellularly within the cytosol and has evolved mechanisms to hijack the actin polymerization machinery leading to actin tail formation and induces the formation of multinucleated giant cells. Among a remarkable number of virulence factors, B. pseudomallei harbours three type III and six type VI secretion systems. The intracellular life cycle and high virulence in rodents, makes B. pseudomallei a promising model pathogen to study basic infection mechanisms. A better understanding of ecological factors determining the environmental dissemination and persistence of B. pseudomallei will be important for understanding the global epidemiology of melioidosis and for undertaking any preventive measures.

Multiresistente Tuberkulose - Bedeutung in Europa Multiresitant Tuberculosis - European Importance

Alexander Indra

Austrian Agency for Health and Food Safety (AGES), Vienna, Austria

alexander.indra@ages.at

Over the last 15 years Austrian tuberculosis cases have steadily decreased, this development was similar in all central European and Scandinavian countries. However one important problem arouses – Multi-Drug-Resistant-Tuberculosis (MDR-TB).

Successful tuberculosis therapy – using at least 4-5 different antimycotic substances – developed after the 2nd World War, was and is dependent on the use of two major substances – Rifampicin and Isoniazid (INH). Resistance to at least both of them leads to prolonged therapy and increase of mortality and is called Multidrug-Resistance-Tuberculosis. Resistance develops by inadequate use of medications during the therapy, which was a major problem during the break-up of the Soviet Union.

MDR-TB patients not only need longer and more expensive antimycotic drugs, it is also noted that these cases infect more people.

In Austria and Central-Europe we have seen also an increase of MDR-TB during the last 15 years, due to our working surveillance- and healthcare-system the transmission to the general population was minimal.

Health of humanitarian workers

Mzia Turashvili

Ärzte ohne Grenzen - Médecins Sans Frontières 1020 Vienna, Austria

*mzia.turashvili@vienna.msf.org

This presentation is about health issues of the humanitarian workers from Western countries who are involved in the humanitarian programmes around the world. We will discuss most common health risks and the ways of their prevention and treatment.

Cloning, expression and molecular characterization of a *Cystoisospora suis* specific uncharacterized merozoites protein

<u>Aruna Shrestha¹</u>^{*}, Nicola Palmieri¹, Ahmed Abd-Elfattah¹, Bärbel Ruttkowski¹, Marc Pagès Bosch², Anja Joachim¹

¹Institute of Parasitology, Department of Pathobiology, University of Veterinary Medicine Vienna, Austria ²HIPRA Laboratorios, Girona, Spain

*Aruna.shrestha@vetmeduni.ac.at

The genome of the apicomplexan parasite Cystoisospora suis (syn. Isospora suis) has recently been sequenced and annotated, opening the possibility for the identification of novel therapeutic targets against cystoisosporosis. It was previously proposed that a 42 kDa uncharacterized merozoites protein encoded by gene CSUI_005805, might be a relevant vaccine candidate due to its high immunogenic score, high expression level and species-specificity. The 1170 bp coding sequence of the CSUI_005805 gene was PCR amplified and cloned into the bacterial expression vector pQE-31. The recombinant CSUI_005805 construct was expressed in Escherichia coli as a (His)₆ tagged fusion protein and purified under denaturing conditions by affinity chromatography using a NiNTA matrix. The specificity of expressed recombinant protein was evaluated in an immunoblot using sera from experimentally infected swine, whereas its localization and level of expression in different developmental stages were determined by quantitative realtime PCR and indirect immunofluorescence assay using polyclonal chicken sera, respectively. The CSUI 005805 gene encodes for a 389 amino acid protein containing a histidine-rich region. Quantitative RT-PCR showed that CSUI_005805 is differentially expressed during the early development of C. suis in vitro, with higher transcript levels in merozoites compared to sporozoites. Interestingly, once the sporozoites invade enterocytes, the transcription level steadily increases throughout the entire merogony indicating that this protein might be important for the survival and establishment of merozoites. Both the recombinant protein and naïve merozoites proteins were specifically recognized by sera from chicken immunized with recombinant CSUI 005805 protein. Moreover, sera of piglets experimentally infected with C. suis also detected the recombinant protein, all of which suggested that despite prokaryotic expression, recombinant CSUI_005805 protein maintained antigenic determinants recognized by protective antibodies against naïve protein and could elicit a immune response in the host. Immunofluorescence and confocal microscopic observations revealed localization primarily at the surface and, occasionally, the inner membrane complex of the parasite.

Immunomodulatory properties of *Toxoplasma gondii* tachyzoites extract are potentially linked to carbohydrate components and lead to suppression of allergic airway inflammation in mice

<u>Mirjana Drinić</u>¹, Angelika Wagner¹, Roman Peschke², Bärbel Ruttkowski², Anja Joachim², Shi Yan³, Iain B.H. Wilson³, Ursula Wiedermann¹ and Irma Schabussova¹

¹Institute of Specific Prophylaxis and Tropical Medicine, Center for Pathophysiology, Infectiology and Immunology, Medical University of Vienna, Vienna, Austria

²Institute of Parasitology, University of Veterinary Medicine, Vienna, Austria ³Department of Chemistry, University for Natural Resources and Life Sciences, Vienna, Austria

We have previously shown that both infection with *Toxoplasma gondii* and treatment with oocysts lysate antigen prevented the development of allergic immune responses and airway inflammation in a mouse model of type I allergy. In this study, we have tested, *in vitro* and *in vivo*, the immunomodulatory properties of extract derived from *T. gondii* tachyzoites, which are rapidly multiplying in infected cells and are associated with acute infection.

Stimulation of splenocytes and bone marrow-derived dendritic cells with tachyzoites lysate antigen (TLA) induced production of pro-inflammatory and regulatory cytokines, such as IFNγ, IL-6 and IL-10. Flow cytometry analysis of these cells showed upregulation of surface markers CD40, CD80, CD86 and MHCII. We could also show that TLA is recognized by TLR2 in transfected HEK cells. Furthermore, immunization of BALB/c mice with TLA induced high IgG1 antibody levels as well as increased levels of pro- and anti-inflammatory cytokines in re-stimulated spleen cell cultures. In a mouse model of allergy, we demonstrate that co-application of TLA during sensitization with OVA reduced development of airway inflammation, reflected in decreased airway hyperresponsivnes and decreased recruitment of eosinophils in bronchoalveolar lavage. Moreover, a significant reduction of TLA curves.

Based on the *in vivo* experiments we further sought to identify which molecules/fractions of TLA exerted the immunomodulatory effects. The TLA extract was heat-inactivated, treated with proteinase K or deglycosylated by means of metaperiodate oxidation in order to determine the role of proteins and glycans. These fractions were tested *in vitro* showing that the immunomodulatory potential of the extract declines upon deglycosylation. Similarly, after chloroform/methanol lipid extraction, fractions will be characterized for their modulatory properties.

Our results contribute to the understanding of the role of parasite molecules in the modulation of the host immune responses and might have an application in the

design of novel vaccines/adjuvants for treatment of immune mediated diseases such as allergy.

Supported by FWF (SFB F46)

Cardiorespiratory parasites in red foxes (*Vulpes vulpes*) from Bosnia and Herzegovina

<u>Adnan Hodžić^{1*}</u>, Amer Alić², Ismar Klebić², Mirsad Kadrić², Emanuele Brianti³, Georg G. Duscher¹

¹Department of Pathobiology, Institut of Parasitology, Universiy of Veterinary Medicine Vienna, Veterinaerplatz 1, 1210 Vienna, Austria

² Department of Pathology, Faculty of Veterinary Medicine, University of Sarajevo, Zmaja od Bosne 90, 71000 Sarajevo, Bosnia and Herzegovina

³ Dipartimento di Scienze Veterinarie, Facoltà di Medicina Veterinaria, Università degli Studi di Messina, Piazza Pugliatti 1, 98168 Messina, Italy

*adnan.hodzic@vetmeduni.ac.at

The aims of the present study were to (*i*) investigate the prevalence and geographical distribution of cardiorespiratory parasites in red foxes, (*ii*) determine genetic diversity of detected parasite species, and (*iii*) to estimate the role of foxes in the transmission cycle to companion animals and humans.

Four species, morphologically and molecularly identified as *Eucoleus boehmi* (64.6%; 51/79), *Eucoleus aerophilus* (69.7%; 154/221), *Crenosoma vulpis* (45.7%; 101/221) and *Linguatula serrata* (1.3%; 1/79) were retrieved from nasal cavity and lungs of 184 (83.3%) animals. *Angiostrongylus vasorum* and *Dirofilaria immitis* was not detected by necropsy or PCR. Furthermore, three distinct haplotypes of *E. aerophilus* (I, III, XV) and two of *C. vulpis* (I, II) previously reported in companion animals and wild carnivores. A new haplotype of *C. vulpis* (named as haplotype V) was also identified based on 12S rRNA gene for the first time.

The present study indicates a high prevalence and wide distribution of cardiorespiratory parasites in fox population in Bosnia and Herzegovina, and supports the existence of transmission patterns between wildlife and pet animals.

References

1. Hodžić A, Alić A, Klebić I, Kadrić M, Brianti E, Duscher GG. Red fox (*Vulpes vulpes*) as a potential reservoir host of cardiorespiratory parasites in Bosnia and Herzegovina. Vet Parasitol. 2016;223:63-70.

Immunology lesson given by "Old Friend" Trichinella spiralis

<u>Alisa Gruden-Movsesijan*</u>, Natasa Ilic, Jelena Cvetkovic, Ivana Mitic, Ljiljana Sofronic-Milosavljevic

Institute for the Application of Nuclear Energy – INEP, University of Belgrade, Belgrade, Serbia

*alisamovsesijan@gmail.com

During evolution the immune system was molded by the influence of an immense variety of stimuli, including helminths. Removal of helminths from everyday environment, due to improved living conditions, could leave the immune system without the "coach". This could result in its dysregulation and consequential development of chronic inflammatory disorders like allergies and autoimmunity. Developed countries reported significant increase in the incidence of autoimmune diseases, which coincided with reduced presence of "old friends", organisms that are necessary for proper development of the immune system. This is the essence of "hygiene hypothesis", strongly supported nowadays by epidemiological, experimental and clinical studies. *Trichinella spiralis* (*T. spiralis*) possess the capacity to create environment that preserves host organism and enables survival of the parasite at the same time. During chronic phase of the infection parasite communicates with the host organism through muscle larvae excretory-secretory products (ES L1). ES L1 products manipulate the immune response of the host not only towards parasitic but also irrelevant antigens (self or non-self in origin). By inducing Th2 and anti-inflammatory responses and activating regulatory network, T. spiralis allows the host to successfully cope with various autoimmune diseases. Indeed, chronic infection with T. spiralis ameliorated the course of experimental autoimmune encephalomyelitis (EAE), animal model of multiple sclerosis, in a dose dependent way. This was accomplished by elevated production of Th2 and regulatory cytokines, IL-4, IL-10 and TGF- β and expansion of regulatory T cells, which suppressed Th1 and Th17 cells responsible for EAE pathogenesis. However, since infection with viable larvae carries risks for the host organism, two different approaches for EAE treatment were created: treatment with cells (T cells isolated from T. spiralis infected animals or dendritic cells stimulated with ES L1 antigens) and treatment with isolated ES L1 antigens. Both treatments mimicked, in a great deal, the immunomodulatory effects of the actual T. spiralis infection and reduced EAE severity in DA rats, although the triggered mechanisms differ. Besides obvious beneficial effects of all applied prophylactic treatments, there are still risks and obstacles to overcome. That is why we need to learn from our "old friends" which molecules and mechanisms are responsible for immunomodulation and to apply that knowledge for human health benefit.

This work was supported by the Ministry of Education, Science and Technological Development, Republic of Serbia, Grant No 173047.

OIE Twinning – a valuable concept to control *Trichinella* infections in Animals and Foodstuff in Tanzania

Karl Schoepf¹*, <u>Walter Glawischnig¹</u>, Henry B. Magwisha³, Godwin A. Minga³, Gianluca Marucci², Edoardo Pozio²

¹Austrian Agency for Health and Food Safety, Innsbruck, 6020, Austria ²Istituto Superiore di Sanità, Rome, 00161, Italy ³Tanzania Veterinary Laboratory Agency, Dar es Salaam, 15487, Tanzania

*karl.schoepf@ages.at

Throughout much of the world, *Trichinella* species have been found to be the causative agents of human trichinellosis, a disease that not only is a public health hazard but also represents an economic problem in animal production and food safety. The World Organization for Animal Health (OIE) has funded a Twinning project on the control of *Trichinella* infections in Animals and Foodstuff of Tanzania. This international partnership with the European Union Reference Laboratory for Parasites (EURLP) as senior partner, the AGES National Reference Laboratory as junior partner and Tanzania Veterinary Laboratory Agency (TVLA) as candidate partner started with a kick off meeting in Dec. 2014 in Rome. The partnership aims at improving the diagnostic competence for controlling of *Trichinella* infection in animals and food products of Tanzania.

Tanzania has approximately 1.6 million pigs and most of them are reared in the rural settings on free range system. Pig rearing provides subsistence income in many families and is an important source of animal protein. Pig production has fast turn over second to poultry providing petty cash to farmers almost all time around the year. Pork consumption is becoming increasingly popular in densely populated towns and cities including Dar es Salaam. Free range management system predisposes pigs to *Trichinella* infection. However, the parasites have not been looked for in slaughtered pigs during meat inspection (lack of lab expertise and financial resources). It is therefore assumed that if *Trichinella* infected pork entering the food chain then a large population of pork consumers would be on risk.

Within this Twinning program competence on diagnosis of *Trichinella* infection in the candidate laboratory will be established under quality assurance standards. An initial survey examining pig carcasses entering the food chain started this year. Further aim is to conduct a risk based survey in wildlife. In sub Saharan Africa, *Trichinella* infections are widespread among wildlife and large predators (lions, hyenas, leopards, etc) are at the apex of *Trichinella* life cycle.

Results from the initial survey will guide decision making whether it is necessary to implement a regular control scheme for pork meat entering the human food chain especially in fast growing urban areas.

The establishment of a Regional Reference Laboratory for *Trichinella* in Dar will put Tanzania into the position to monitor *Trichinella* infections in domestic and wild animals. The production of *Trichinella* - free animals and derived food products are essential regarding food safety issues. When considering trade with neighbouring countries it is essential to meet food safety standards.

Prevalence of insecticide resistance house flies (*Musca domestica*) on pig farms in Schleswig-Holstein, Germany

S. Steuber¹*, J. Hildebrand², S. Reissert², K. Sievert³, B. Bauer², P.-H. Clausen²

¹Federal Office of Consumer Protection and Food Safety, 10117 Berlin, Germany ²Institute for Parasitology and Tropical Veterinary Medicine, Freie Universität Berlin, 14163 Berlin, Germany ³Syngenta Crop Protection AG, 4058 Basel, Switzerland

*stephan.steuber@online.de

Disturbance by house flies (*Musca domestica*) is considered a major economic problem in animal husbandry. Therefore, fly control constitutes an integral part of livestock management. However, non-strategic use of insecticides might lead to rapid buildup of insecticide resistance (IR).

The susceptibility of *M. domestica* against deltamethrin (DTM) was assessed on 40 farms using the FlyBox[®]-test (280mg DTM/m²). The field populations were further tested by a feeding assay against neonicotinoids (thiamethoxam, imidacloprid) and azamethiphos (phosphoric ester). Insect growth regulators (cyromazine, triflumuron) were additionally evaluated. Subsequently, 17 fly populations were lab-reared and their offspring exposed to topical applications of pyrethrum, DTM, azamethiphos, thiamethoxam and imidacloprid.

Resistance (mortality rates \leq 90%) to DTM were observed in 85% of the fly populations. When exposed to thiamethoxam, 13 populations (33%) revealed moderate resistance (mortality rate 40-89%). Only 4 populations (10%) displayed 100% paralysis when imidacloprid was applied. Ten strains (25%) were sensitive against azamethiphos, the majority (75%) showing a moderate degree (\leq 90%) of resistance. Cyromazine proved highly effective (100% inhibition of larval development), whereas 30% of the fly populations emerged from the triflumuron treated culture medium at the recommended concentration of 5 mg/kg.

In the laboratory only 1 strain was susceptible to pyrethrum at the discriminating dose (DD) of 2.2 μ g/fly. Six strains tested showed high DTM resistance. Resistance was confirmed against azamethiphos, 15 strains showed moderate to high resistance at 0.32 μ g/fly. The majority of the populations (59%) was also highly resistant against thiamethoxam (DD of 0.32 μ g/fly) and imidacloprid (41% at 2 μ g/fly).

Insecticide use has to be considered as last resort for managing house flies or other insect pests. Potential occurrence of IR should be evaluated before using insecticides. Rotation of active ingredients, frequent manure removal and biological means of control may delay IR development.

Parasites of the Alpine Ibex (*Capra i. ibex*) in North Tyrol – faunal diversity and abundance

S. Rehbein¹, C. Messner², M. Visser¹

¹ Merial GmbH, Kathrinenhof Research Center, Rohrdorf, Germany

² Tierarztpraxis Mag. C. Messner, Schwaz, Austria

The Alpine ibex was almost extinct at the beginning of the 19th century. In the territory of the today's Tyrol (and Austria), last ibex, which were reported from the Floite and Gunggl valleys of the Zillertal mountains, disappeared in the early 18th century. The first successful release of ibex in Austria was initiated in Blühnbach, Salzburg in 1924. In the Tyrol, first releases of ibex occurred in the early 1950s including the successful re-introduction in the Pitz valley of the Ötztal Alps. Currently, more than 30 ibex colonies exist in North Tyrol with a total population of about 5000 head.

As no studies on the parasite fauna of the ibex from North Tyrol have been reported before, viscera or carcasses of 32 ibex originating from 7 colonies were examined with standard techniques during the years of 2008 to 2013.

Gastrointestinal helminths (total 32 species [morphs for Ostertagiinae] of nematodes [abomasum - 13, small intestine - 14, large intestine - 5] and *Avitellina centripunctata* and *Moniezia* spp. cestodes) and lungworms (total 5 species of protostrongylids and *Dictyocaulus* sp.) were recovered from all ibex. In addition, *Taenia hydatigena* metacestodes were recorded in 12.5% of the ibex and examination of heart and/or diaphragmatic muscle samples revealed sarcocysts in 65.4% ibex. No parasites were found in the livers. As regards ectoparasites, sarcoptic mange mites, biting lice (*Holakartikos crassipes*) and keds (*Melophagus rupicaprinus*) were isolated from the ibex.

Individual ibex harboured 3 to 11 species of nematodes in the abomasum, 2 to 7 species of nematodes in the small intestine and 0 to 4 species of nematodes in the large intestine. Twelve species of gastrointestinal nematodes and 2 species of protostrongylid lungworms were recorded from more than 60% of the ibex. In one ibex, up to 20 or 4 species of nematodes were recovered from the gastrointestinal tract or lungs, respectively.

Individual nematode burden of the gastrointestinal tract and lungs ranged from 260 to 22349 (geometric mean, 5828) and 16 to 1694 (geometric mean, 157), respectively.

Aiming to obtain basic parasitological data for the ibex in North Tyrol, this study demonstrated a high diversity of parasites and a remarkable parasite burden in the ibex.

Dirofilariosis, an overview and a multidisciplinary approach conducted in an European country

Ana Margarida Alho¹, Luís Madeira de Carvalho¹

¹CIISA, Faculty of Veterinary Medicine, ULisboa, Lisboa, Portugal

*margaridaalho@fmv.ulisboa.pt

Cardiopulmonary dirofilariosis caused by Dirofilaria immitis, is a zoonotic mosquito-borne disease increasingly reported worldwide, with a potentially severe outcome in companion animals [1]. Several factors have been linked to its expansion, namely faster and incremented global transport and international commerce, urbanization and abundance of wildlife hosts, demographic and political changes, drug resistance and climate alterations [2]. Here, a systematic review is presented as well as a map of the current distribution of Dirofilaria in using geospatial tools. Concurrently, the authors describe Europe. а multidisciplinary study conducted in Portugal, involving: a) an epidemiological survey to investigate the prevalence of *D. immitis* in dogs and red foxes; the report of an outbreak of *D. immitis* in pinniped species and the description of a new host; b) a degree day model to estimate the transmission risk of Dirofilaria larvae between mosquito and reservoirs based on air temperature [3]; c) an alternative method for mechanical removal of *D. immitis* in dogs using a homemade snare [4]; and d) a survey to assess owner's awareness and public perception of parasitic diseases and parasite control practices [5].

Overall, the current data show that dirofilariosis is a prevalent disease, widely distributed in many European countries. Additionally, data show the urgent need to raise awareness among general population regarding parasite transmission, treatment and regular prophylaxis. Large epidemiological studies to estimate the occurrence of *D. immitis* infections in domestic and wild animals, coupled with continued entomological surveillance programs are needed to improve scientific knowledge and to prevent the spread into non-endemic areas, therefore limiting potential zoonotic outbreaks. An integrated approach under the scope of "One World, One Health" at local and global scale is vital to control this pathogen and promote higher animal and public health standards.

- 1. Simón F, Siles-Lucas M, Morchón R, González-Miguel J, Mellado I, Carretón E, et al. Human and animal dirofilariasis: the emergence of a zoonotic mosaic. Clin Microbiol Rev. 2012;25:507-44.
- 2. Colwell DD, Dantas-Torres F, Otranto D. Vector-borne parasitic zoonoses: emerging scenarios and new perspectives. Vet Parasitol. 2011;182:14–21
- Alho AM, Nunes T, Rinaldi L, Meireles J, Belo S, Deplazes P, Madeira de Carvalho L. Transmission risk of dirofilariosis in Portugal. Parasit Vectors. 2014; 7 (Suppl 1):O16.
- Alho AM, Fiarresga A, Landum M, Lima C, Gamboa Ó, Meireles J, Sales Luís J, Madeira de Carvalho L. A Homemade snare: An alternative method for mechanical removal of *Dirofilaria immitis* in dogs. Vet Med Int. 2016; 5780408.
- Matos M, Alho AM, Owen SP, Nunes T, Madeira de Carvalho L. Parasite control practices and public perception of parasitic diseases: A survey of dog and cat owners. Prev Vet Med. 2015;122(1-2):174-80.

Filarioid helminths in mosquitoes from the Danube Delta/Romania and the analysis of these vectors for potential vector competence

<u>Victoria Wimmer</u>¹, Angela Monica Ionică², Carina Zittra¹, Natascha Leitner¹, Jan Votýpka³, David Modrý^{4,5}, Andrei Daniel Mihalca², Katja Silbermayr¹, Hans-Peter Fuehrer^{1*}

*hans-peter.fuehrer@vetmeduni.ac.at

¹ Department of Pathobiology, Institute of Parasitology, University of Veterinary Medicine Vienna, 1210 Vienna, Austria

² Department of Parasitology and Parasitic Diseases, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine, Calea Mănăştur 3-5, Cluj-Napoca, Romania

³ Department of Parasitology, Faculty of Sciences, Charles University, Viničná 7, 12844 Prague, Czech Republic

 ⁴ Department of Pathology and Parasitology, Faculty of Veterinary Medicine, University of Veterinary and Pharmaceutical Sciences, Palackého tr. 1946/1, 612
42 Brno, Czech Republic

⁵ Biology Centre, Institute of Parasitology, Czech Academy of Sciences, Branisovska 31

370 05 České Budějovice, Czech Republic

In the past decades both *Dirofilaria immitis* and *D. repens* have spread from historically endemic areas to central and eastern European countries. Several studies have shown that *Dirofilaria* species are present in the southern and southeastern areas of Romania. However, information about the vectors in the Danube Delta and their vector competence is lacking.

In July 2015 more than 5,000 mosquitoes were collected in the Danube Delta in Romania at various locations (including mosquito traps next to a dog infected with both *D. immits* and *D. repens*). Mosquitoes were classified to species-level using the key after Becker et al.. In one part of the study specified mosquitoes were pooled (up to 25 individuals per day/trap/mosquito species). DNA was extracted and the samples were screened for filarioid helminths using conventional PCRs. For the second part of the study 300 specified mosquito individuals caught at the trap next to a microfilariaemic dog positive for *D. immitis* and *D. repens* were segregated into head/thorax and abdomen prior to DNA extraction. Each thorax/head and abdomen was screened for the presence of filarioid DNA separately. All positive PCR products were further analysed by sequencing. The aim for the third part of the study was to establish a novel qPCR detecting *D. repens*, *D. immitis* and *Acanthocheilonema reconditum*.

Mosquitoes were sampled within the training school of WG1 under the frame of EurNegVec COST Action TD1303. Parts of this study were funded by the ERA-Net BiodivERsA, with the national funders FWF I-1437, ANR-13-EBID-0007-01 and DFG BiodivERsA KL 2087/6-1 as part of the 2012-13 BiodivERsA call for research proposals.

Molecular epidemiology of mosquito-borne diseases in Eastern Austria

<u>Übleis S. S.¹</u>, Cuk C.¹, Nawratil M.¹, Berer D.¹, Eigner B.¹, Zittra C.¹, Obwaller A.G.², Lebl K.³, Zechmeister T.⁴, Lamp B.⁵, Brandt S.⁶, Roiz D.⁷, Fuehrer H.-P.^{1*}

¹Institute of Parasitology, Department of Pathobiology, University of Veterinary Medicine Vienna, Vienna, Austria

²Federal Ministry of Defence and Sports, Division of Science, Research and Development, Vienna, Austria

³Institute for Veterinary Public Health, University of Veterinary Medicine Vienna, Vienna, Austria

⁴Biological Station Lake Neusiedl, Burgenland, Austria

⁵Institute of Virology, Department of Pathobiology, University of Veterinary Medicine Vienna, Vienna, Austria

⁶ Research Group Oncology, Equine Clinic, Department of Companion Animals and Horses, University of Veterinary Medicine, Austria

⁷MIVEGEC (Infectious Diseases and Vectors: Ecology, Genetics, Evolution and Control), UMR IRD224-CNRS5290-UM, Institut de Recherche pour le Développement (IRD), Montpellier, France

*hans-peter.fuehrer@vetmeduni.ac.at

Global warming and rising globalization have essential effects on the distribution and circulation areas of insect vectors (e.g. mosquitoes) and their associated pathogens (e.g. *Dirofilaria*, West-Nile virus). Especially in Central Europe high temperatures during the summer months coupled with exceedingly mild winters, favor the introduction and spread of non-indigenous species. A range of bacterial, parasitic and viral agents are transmitted via this insect route and have the potential of causing exotic diseases in previously unprecedented areas. Permanent establishment of new competent vector species lead to an increased risk of endemic vector-borne infections. Therefore, information on the distribution patterns of mosquitoes and their transfer function as disease vectors is crucial and with utmost importance for public health, veterinary medicine and scientists.

Still, there is markedly few data on the pathogen load transmitted by mosquitoes in Metropolitan Vienna, Lower Austria and Burgenland. During this ERA-Net BiodivERsA project almost 30,000 mosquitoes were collected in Vienna, Lower Austria and Burgenland within the investigation periods of the years 2014 and 2015. Due to morphological standard characteristics, female mosquitoes were classified to species-level and pooled up to 50 individuals according to their date of collection and sampling site. Until further examinations the pools were stored at -80°C. Nucleic acids were extracted and screened for the presence of DNA of *Francisella tularensis* and filarioid helminths as well as genomic RNA of flaviviruses by using molecular techniques. This approach provides results of the pathogen screening and the occurrence of potentially infectious agents transmitted by mosquitoes in the examined regions during 2014 and 2015.

This study was funded by the ERA-Net BiodivERsA, with the national funders FWF I-1437, ANR-13-EBID-0007-01 and DFG BiodivERsA KL 2087/6-1 as part of the 2012-13 BiodivERsA call for research proposals.

Spatiotemporal mosquito species distribution in Eastern Austria driven by environmental parameters - final results

<u>Carina Zittra¹</u>, Simon Vitecek², Adelheid G. Obwaller³, Heidemarie Rossiter⁴, Barbara Eigner¹, Thomas Zechmeister⁵, Johann Waringer², Hans-Peter Fuehrer^{1*}

¹ Institute of Parasitology, Department of Pathology, University of Veterinary Medicine, Vienna 1210, Austria

² Department of Limnology and Bio-Oceanography, University of Vienna, Vienna, Austria

³ Federal Ministry of Defence and Sports, Division Science, Research and Development, Vienna, Austria

⁴ Donaustraße 73, 3421 Höflein/Donau, Austria

⁵ Biological Station Lake Neusiedl, Burgenland, Austria

*hans-peter.fuehrer@vetmeduni.ac.at

Dispersal and establishment of native and non-indigenous mosquito species as well as emerging vector-borne diseases are mainly driven by international trade and global change. Thus, data on mosquito ecology, in particular seasonal and spatial distribution patterns of mosquito communities is crucial for the understanding of vector-pathogen dynamics in Europe. However, basic information on the mosquito species inventory, its genetic diversity, and seasonal and spatial variations of mosquito communities in Austria is still insufficient. In our study, female mosquitoes were monitored at 35 permanent and 23 non-permanent trapping sites using carbon dioxide baited traps twice a month from April to October 2014 and 2015. Possible effects of environmental factors on seasonal and spatial mosquito distribution were analysed using multivariate statistical methods; differences in the proportional mosquito abundance were identified using likelihood ratio tests (G-tests) of goodness of fit. Furthermore a canonical correspondence analysis (CCA) was used to relate mosquito community fluctuations to variability of environmental parameters. Potential effects of environmental parameters possibly affecting mosquito community composition using permutational multivariate assessed analysis of variance were (PERMANOVA) on dissimilarity matrices; environmental parameters with a significant contribution to the observed patterns were modelled in ordination space to estimate non-linear relationships.

In this study, 29,735 specimens were sampled and 26 of 42 native as well as two of four non-native mosquito species were reconfirmed in Eastern Austria.

Statistical analyses revealed significant differences in mosquito abundance between sampling years and provinces due to environmental variables and detected environmental parameters influencing spatial and temporal mosquito species distribution.

We present effects of climatic parameters and habitat structure on mosquito communities in Eastern Austria and thus deliver highly relevant baseline data for future mosquito prediction models and mosquito surveillance programmes.

Parts of this research were funded by the ERA-Net BiodivERsA, with the national funders FWF I-1437, ANR-13-EBID-0007-01 and DFG BiodivERsA KL 2087/6-1 as part of the 2012-13 BiodivERsA call for research proposals.

Morphological and molecular identification of *Synhimantus (Synhimantus) laticeps* (Rudolphi, 1819) Railliet, Henry et Sisoff, 1912 (Nematoda, Acuariidae) from the barn owl (*Tyto alba*) and the common kestrel (*Falco tinnunculus*) in Austria

<u>David Ebmer¹</u>^{*}, Barbara Eigner¹, Hans-Peter Fuehrer¹, Helmut Sattmann², Anja Joachim¹

 ¹ Institute of Parasitology, Department of Pathobiology, University of Veterinary Medicine Vienna, Veterinaerplatz 1, 1210 Vienna, Austria
² Third Zoological Department, Natural History Museum Vienna, Burgring 7, 1010 Vienna, Austria

*david.ebmer@vetmeduni.ac.at

In the framework of the biodiversity initiative and barcoding project "Austrian Barcode of Life" (ABOL) post mortem examinations of the gastro-intestinal tracts of different species of wild birds were carried out and several adult helminths could be retrieved. In the gizzard of two barn owls (*Tyto alba*) and one common kestrel (*Falco tinnuculus*) acuariid nematodes belonging to the species *Synhimantus* (*Synhimantus*) laticeps (Rudolphi, 1819) Railliet, Henry et Sisoff, 1912 were discovered.

Morphological identification of this parasitic nematode by morphometric comparison and scanning electron microscopic photographs was accomplished. Furthermore, genetic identification of individual parasites based on a fragment of the mitochondrial *cytochrome c oxidase subunit I* (COI) gene was carried out. Our work constitutes the first COI-based DNA barcoding of *Synhimantus (S.) laticeps* and its first description in the barn owl (*Tyto alba*) in Austria.

Novel approaches to diagnosing malaria. Development and validation of a High Resolution Melting Curve Analysis assay for human malaria diagnosis

Aline Lamien-Meda^{1*}, Hans-Peter Fuehrer², David Leitsch¹, Harald Noedl¹

¹Institute for Specific Prophylaxis and Tropical Medicine, Medical University of A-1090 Vienna, Austria

²Institute of Parasitology, University of Veterinary Medicine, Vienna, Austria

*Aline.LamienMeda@meduniwien.ac.at

A major step-up in worldwide malaria control efforts in the past decades has resulted in a significant reduction of mortality and clinical episodes in most malaria-endemic countries. At the same time asymptomatic infections have gained importance as a reservoir of new infections and epidemics. Novel, more sensitive tools are therefore urgently needed to replace microscopy as the gold-standard of diagnosis also in resource-limited environments.

Since the development of PCR based methods in malaria diagnosis in the late 1980's, several methods targeting the plasmodium 18S SSU RNA gene have been developed and widely deployed. These methods include conventional nested and semi nested PCR, Lamp PCR and real time PCR [1, 2]. However, the plasmodium species-specific identification by these methods requires multiplexing or several time consuming steps using primer pairs that are specific to each of the plasmodium species.

High Resolution Melting (HRM) curve analysis is a simple and fast post-PCR analysis which has been successfully applied for genotyping, including pathogentyping. In this procedure, the region of interest is amplified in the presence of a specialized dsDNA binding dye and a gradual denaturation of the amplicons produces characteristic melting profiles. Recently, a HRM assay targeting the rSSU RNA was described for the simultaneous detection and typing the five plasmodium species affecting human [3]. The adoption of such HRM assay would help saving time in plasmodium species identification.

We describe the development of a real time PCR-HRM assay for the first time targeting the mitochondrial DNA for the simultaneous detection, species specific identification and quantification of *P. falciparum*, *P. vivax*, *P. knowlesi*, *P. malariae*, *P. ovale curtisi* and *P. ovale wallikeri*, with high specificity and sensitivity. This method combines several advantages of targeting the mitochondrial DNA, namely that it is more conserved within each of the plasmodium species while at the same time being present in multiple copies within each parasite.

References

- 1. Fuehrer H-P, Noedl H. Recent Advances in Detection of *Plasmodium ovale*: Implications of Separation into the Two Species *Plasmodium ovale wallikeri* and *Plasmodium ovale curtisi*. JCM, 2014, 52, 387-391.
- Alemayehu S, Feghali KC, Cowden J, Komisar J, Ockenhouse CF, and Kamau E. Comparative evaluation of published real-time PCR assays for the detection of malaria following MIQE guidelines. Malar J, 2013, 12,277-285.
- Chua KH, Lim SC, Ng CC, Lee PC, Lim YAL, Lau TP, Chai HC. Development of high resolution melting analysis for the diagnosis of human malaria. Sci Rep. 2015, 5, 15671-84.

Molecular genetics and morphological identification of Austrian Acanthocephala (Kohlreuther, 1771)

Susanne Reier^{1*}, Elisabeth Haring¹, Hans-Peter Führer², Thomas Schwaha³, Helmut Sattmann⁴

¹ Natural History Museum Vienna, Central Research Laboratories, Burgring 7, 1010 Vienna, Austria

² Institute of Parasitology, Department of Pathobiology, University of Veterinary Medicine Vienna, Veterinaerplatz 1, 1210 Vienna, Austria

³ Department of Integrative Zoology, University of Vienna, Althanstraße 14, 1090 Vienna, Austria

⁴ Natural History Museum Vienna, 3rd Zoological Department, Burgring 7, 1010 Vienna, Austria

*reier_susanne@yahoo.de

Although Acanthocephala are a well-studied phylum, molecular genetic methods for species identification have been rarely applied in this taxon. Genetic characterisation of species by DNA barcoding is gaining increasing importance, especially in a large and highly diverse group of often small sized animals like Acanthocephala. The purpose of this study was a first survey of fish-parasitizing Acanthocephala in Austria based on DNA barcoding. The aim was to test various primer sequences, to register species occurrences in different aquatic systems and host species, and to get insights into intra- and interspecific diversities. Here we report our first results.

In 26 different fish species from various rivers in Styria, Vienna, Upper Austria and Lower Austria we found 307 acanthocephalans. For molecular genetic identification the DNA barcoding sequence, a section of the *cytochrome oxidase subunit 1* gene (*COI*) was analysed. Various primers, mostly specific ones designed for the phylum Acanthocephala, were used. In addition, morphological identification was conducted by different staining and microscope methods such as light microscopy, scanning electron microscopy and confocal laser scanning microscopy as a novel method for species identification in Acanthocephala.

Three genera of Acanthocephala could be detected: *Acanthocephalus* (Koehlreuther 1771), *Echinorhynchus* (Zoega in Müller 1776) and *Pomphorhynchus* (Monticelli 1905). All genera belong to the class Palaeacanthocephala. In general the morphological determination was in accordance with the genetic assignment.

Within the genus *Pomphorhynchus* three species could be differentiated: *P. tereticollis*, *P. laevis* and a mitochondrial lineage that could not be assigned to any species. The average genetic distance between this lineage and the *P. laevis*-sequences (from our own analysis as well as from Genbank) amounts to 10.2% (p distance). The divergence between this unidentified species and *P. tereticollis* even was 21.1%. There were only faint morphological differences between the three species, e.g., projections on the posterior row of hook plates. The Austrian *P. laevis* were larval stages and consequently could not be identified morphologically. Therefore, the identification was only possible through DNA

barcoding. The average distances between our two *P. laevis* samples and published sequences were very low (~0.7%).

Our results demonstrate that DNA barcoding is suitable to identify Acanthocephala, because of their low anatomical diversity. This pilot study also revealed how important it is to link morphological and genetic characteristics, particularly because of the lack of reference sequences in GenBank and the risk of species misidentification.

STD – Virusinfektionen

Heimo Lagler, MPH

War früher bei Virusinfektionen ein therapeutischer Nihilismus ausgeprägt, hat sich dies in den letzten Jahren entscheidend gewandelt. So stehen heute nicht nur antivirale Substanzen gegen *Herpes simplex* -, *Herpes zoster* - oder Zytomegalie - Virusinfektionen zur Verfügung, sondern auch Virostatika gegen HIV, Hepatitis B und C zur Verfügung.

Neu überarbeiteter Ratgeber f. Ärzte zu Skabies

Anton Aebischer

Fachgebiet 16, Robert Koch-Institut, 13353 Berlin, DE

AebischerA@rki.de

Die Herausgabe der Ratgeberreihe durch das Robert Koch-Institut erfolgt auf der Grundlage des § 4 des in Deutschland geltenden Infektionsschutzgesetzes. Praktisch bedeutsame Angaben zu wichtigen Infektionskrankheiten sollen aktuell und konzentriert der Orientierung dienen. Die Beiträge werden in Zusammenarbeit mit den Nationalen Referenzzentren, Konsiliarlaboren sowie weiteren Experten erarbeitet. Die Erstpublikation und deutlich überarbeitete Folgeversionen werden im Epidemiologischen Bulletin und im Internet (<u>www.rki.de/ratgeber</u>) veröffentlicht.

Die Überarbeitung des Ratgebers Skabies und der S-1 Leitlinie der Deutschen Gesellschaft für Dermatologie zu Skabies wurde durch die hohe Aufmerksamkeit, die die Erkrankung aufgrund der Flüchtlingswelle 2015 erhielt, notwendig. Im Vortrag wird auf die wichtigsten Neuerungen eingegangen und die Grundlagen für die Empfehlungen vorgestellt. Darüber hinaus werden Ergebnisse einer die Überarbeitung des Ratgebers begleitenden Erhebung zum Vorkommen der Skabies und daraus resultierender Probleme und Belastungen des Öffentlichen Gesundheitsdienstes vorgestellt.

Filzläuse (pubic lice, scrab lice)

Herbert Auer

Institute for Specific Prophylaxis and Tropical Medicine, Center of Pathophysiology, Infectiology and Immunology, Medical University Vienna

Phthirus pubis, the pubic or scrab louse is considered as one of the man's closest companion, which still occurs throughout the whole world and which is transmitted mainly by sexual contacts. This lecture gives an overview about biology, diagnosis, nosology, treatment and management of *Phthirus pubis*.

Military and Malaria in the last century: Epidemics, Deployments and Drug Development.

G Dennis Shanks

Australian Army Malaria Institute and School of Population Health, Univ. Queensland COL US Army (retired)

Malaria is one of the few infections that can decide military campaigns due to its ability to cause mass casualties in soldiers deployed to the tropics. During the Second World War alone malaria was the defining element of medical geography during fighting in the Philippines, Burma and the Southwest Pacific Islands. Although there have been attempts to utilize malaria against invaders, the usual situation is an epidemic in non-immune soldiers which is initiated and sustained by asymptomatic infections in the indigenous population. Drug resistance was the main factor in malaria during the Vietnam War which was addressed by parallel drug development efforts by the US and Chinese Armies yielding mefloquine and artemisinin respectively. During recent campaigns in Somalia and Afghanistan, relapsing malaria after leaving the endemic area was the problem faced by military physicians. Interventions into West Africa carry a very high risk of falciparum malaria which has severely effected some national elements but not when strict chemoprophylaxis compliance was possible as during the recent Ebola epidemic. United Nations missions have particular challenges due to the mixed nature of the national military groups and differing levels of medical support. The currently used chemoprophylactic drugs (atovaquone/proguanil, doxycycline, mefloquine) are all administered effective if regularly under military discipline. New chemoprophylactic regimens are being sought that are easier to conduct directly observed therapy in large numbers of deployed soldiers.

Malaria – Past, Present and Future

Harald Noedl

Institute of Specific Prophylaxis und Tropical Medicine, Medical University of Vienna, Vienna, Austria

harald.noedl@meduniwien.ac.at

Throughout human history malaria has always been closely tied to human and cultural development, not just in the tropics but also in the temperate climate of Central Europe. Since 1901 four Nobel Prize awards were directly linked to malaria, covering a wide range of discoveries that have ultimately led to malaria becoming a curable disease. However, new challenges in the control and elimination of malaria and the emergence of drug resistance to virtually all currently available antimalarials is threatening many of the achievements of the past decades. Since 2000 virtually all malaria-endemic countries have officially adopted artemisinin-based combination therapies as first or second line therapy for the treatment of P. falciparum malaria. Now artemisinin resistance is about to add another level of complexity to the challenge of malaria control and elimination. New drugs and potentially vaccines are therefore urgently needed to replace or improve current treatment strategies.

Cerebral malaria – adjunctive therapies are essential

Erich Schmutzhard

Department of Neurology, Neuro-Critical Care Unit, Medical University Innsbruck, Austria

erich.schmutzhard@i-med.ac.at

Plasmodium falciparum, the only cause of cerebral malaria (CM), still kills – worldwide - up to 440.000 patients, mainly children and non-immune travellers per year (WHO, 2015). In most cases CM is the leading part of a multi-organ disease, the major pathophysiological mechanism being impairment of microcirculation, through a wide variety of pathways. Steadily growing knowledge of these pathopysiological processes changed the attitude towards invasive therapeutic invasiveness and monitoring methods.

Earliest possible diagnosis and initiation of intravenous Artesunate is the prerequisite of survival. Stabilisation of the cardiopulmonary system, i.e. sufficient fluid administration and, if necessary of catecholamines allows improvement of organ-perfusion, including kidney, liver and brain. Diffuse brain edema, mainly of the vasogenic type, is the underlying pathophysiological mechanism which eventually leads to increased intracerebral pressure (ICP) and, hence decreased cerebral perfusion pressure (CPP). Both, dexamethasone and continuous osmotherapy – in prospective randomized trials - have been shown NOT to improve survival, they are even adding to morbidity. The mechanisms for these failing therapeutic options will be discussed. Prophylactic anticonvulsive therapy, equally, deteriorates the prognosis.

Every patient suffering from CM needs

-emergency admission to a (N)ICU,

-organ replacement therapy (kidney, coagulation etc)

-cautious, at the best, neuroprotective improvement of CPP, choosing the correct therapeutic strategies,

-optimizing blood glucose and thereby relieving the metabolic stress in the brain parenchyma,

-optimizing the ventilation and avoiding

---hypocapnia which leads to intracranial vasoconstriction and thereby to additional cerebral hypoxia and metabolic distress or

---hypercapnia which leads to intracranial vasodilatation and thereby to increased intracranial volume and, consequently, increased ICP and decreased CPP and -maintaining normoxia

-in case of epileptic seizures barbiturates should be avoided, but anticonvulsive therapy immediately initiated, at the best with benzodiazepines, midazolam or ketamine.

-The intestine is similarly affected by the disturbance of microcirculation; therefore hematogenic translocation of intra-intestinal pathogens (e.g. Gram-negatives) may occur and provoke a fulminant sepsis syndrome.

Children with hearing loss secondary to severe and cerebral malaria do not regain full cochlear function after four years – a follow-up study.

Elisa Reiterer^{1,4}, Erich Schmutzhard¹, Akim Adegnika², Peter Kremsner^{2,3}, Joachim Schmutzhard⁴

¹Department of Neurology, Medical University Innsbruck

²Centre des Recherches Médicales de Lambaréné, Hôpital Albert Schweitzer, Lambaréné, Gabun

³Department of Tropical Medicine, Travel Medicine and Human Parasitology,

University of Tübingen

⁴Department of Oto-Rhino-Laryngology, Medical University Innsbruck

It has been shown that severe and cerebral malaria provoke acute cochlear malfunction in children, demonstrated by a decrease of transitory Otoacoustic Emissions (tOAE) correlations¹. This study aims to determine whether cochlear hearing loss persists for 4 years after recovery.

Follow-up tOAEs were performed on site (CERMEL, Hôpital Albert Schweitzer, Lambaréné, Gabun); 33 out of 90 original participants of our previous study could be retrieved and were re-examined, 32 could be included (in one case the identity of the participant could not be verified)¹. Of the 57 missing participants, 51 could not be contacted, 1 had moved away, 4 refused to cooperate, and 1 had died.

The cohort had a mean age of 8.97 years, 14 were female, 18 male. 31 had been originally admitted with severe, 1 with cerebral malaria. 84.8% of participants presented with a tOAE correlation rate of >60% on both ears (the cut-off for good cochlear function); in the control group, 92.52% passed tOAE examination on both ears. Recurrent severe malaria was associated with a worse tOAE correlation rate. The only patient with cerebral malaria did not present with a worse outcome. Age at infection and sex had no influence.

A slight cochlear malfunction is persistent as the overall outcome was good but hearing was still significantly worse in the original severe malaria patients than in the healthy control population. Hearing loss (tOAE <60%) persisted in 5 out of 32 patients on at least one ear; still, the severity of the initial tOAE-decrease did not correlate with a worse outcome.

1 Schmutzhard, J., Lackner, P., Helbok, R. *et al.* Severe malaria in children leads to a significant impairment of transitory otoacoustic emissions--a prospective multicenter cohort study.*BMC Med.* (2015); **13**: 125.

EBOLA: A personal experience report based on an EMLab deployment to western Africa

Rupert Bliem

Armament and Defence Technology Agency, Austrian Armed Forces, Austria

rupert.bliem@bmlvs.gv.at

Ebola is a viral hemorrhagic fever of humans and other primates caused by ebolaviruses. The disease has a high case fatality rate up to 90 percent. The latest outbreak in West Africa, which occurred from December 2013 to January 2016 was the reason for a mobile laboratory deployment designated as EMLab.

The European mobile laboratory was developed by Bundeswehr Institute of Microbiology in Munich, the whole project is coordinated by the Bernhard-Nocht-Institute for Tropical Medicine in Hamburg and funded by the EU.

The author will give a brief overview about his daily laboratory routine under African circumstances.

Reisemedizin: Aufgaben und Herausforderungen im Wandel der Zeit

Robert Steffen

Institut für Epidemiologie, Biostatistik und Prävention, WHO Collaborating Centre for Traveller's Health, Universität Zürich — Zürich, Schweiz

robert.steffen@uzh.ch

Nach dem 2. Weltkrieg ermöglichten Wirtschaftswunder und schnellere Flugzeuge der breiten Bevölkerung zunehmend ferne Ziele aufzusuchen. Für Destinationen in den Tropen und Subtropen, wo die hygienischen Bedingungen oft desolat waren, gab es in tropenmedizinischen Fachbüchern zwar Empfehlungen zu Impfungen und zur Prophylaxe der Malaria aber diese waren widersprüchlich. Denn sie beruhten auf traditionellen Expertenmeinungen; es fehlte die Evidenz. Diesen Mangel erkannten diverse Forscher — in Österreich bereits in den 70er Jahren G. Wiedermann, F. Ambrosch und etwas später H. Kollaritsch — es wurden sowohl epidemiologische Daten zu Gesundheitsrisiken erhoben, wie auch die Wirksamkeit und Nebenwirkungen prophylaktischer Maßnahmen dokumentiert.

Damit war die Reisemedizin geboren. Sie konzentrierte sich zunächst auf *pretravel* (gemäß WHO *Travel Health*), d.h. Empfehlungen zu Impfungen, Malariaprophylaxe, Reisedurchfall. Aufgabe war und bleibt, möglichst alle Reisenden bei guter Gesundheit zu bewahren, beziehungsweise im Falle von gewissen Krankheiten eine Notfall-Selbsttherapie zu ermöglichen. Später wurden in einem Schulterschluss mit den Tropenmedizinern und Infektiologen auch *posttravel* Konsultationen in die *Travel Medicine* eingeschlossen.

Die Reisemedizin ist voller Dynamik, es ergeben sich immer wieder neue Herausforderungen. Vor allem verändert sich die epidemiologische Lage, gewisse Risiken nehmen dank verbesserter Infrastruktur in den Zielländern ab, neue Seuchen zwingen uns blitzschnell eine Expositionsprophylaxe zu entwickeln, bis allenfalls Vakzinen zur Verfügung stehen, oder auch nicht. Nebenwirkungen von Malaria Medikamenten und selten auch von Impfstoffen zwingen uns die Strategien zu überdenken. Resistente Pathogene und Folgeschäden der Reisediarrhöe veranlassen uns den verbreiteten Gebrauch von Antibiotika zu reduzieren. Der Medizinal-Tourismus resultiert ebenfalls in vermehrtem Import von multiresistenten Keimen, was lässt sich dagegen vorkehren? Am Horizont stehen wohl billigere Optionen zur Vorbeugung gegen die Tollwut. Unbefriedigend ist die Tatsache, dass ein wesentlicher Anteil von Reisenden, vor allem auch die einem hohen Risiko ausgesetzten VFRs (visiting friends and relatives) keinerlei reisemedizinische Empfehlungen einholt. Zunehmend aber sind wir gefordert durch neue Typen von Reisenden: Migranten und Flüchtlinge. Diese verdienen nicht nur aus humanitären Gründen eine optimale Betreuung, letztlich schützen wir damit auch unsere eigene Bevölkerung.

Update Weltseuchenlage

Herwig Kollaritsch

Zentrum f.Reisemedizin, Wien

Neu oder wieder auftretende Infektionskrankheiten erregen ganz besonders das mediale Interesse und dadurch werden Reisende sehr oft in ihrer Reisewahl oder überhaupt in ihrer Reiseentscheidung nachhaltig beeinflusst. In diesem Vortrag wird eine Übersicht über die derzeit wesentlichen epidemiologischen Entwicklungen weltweit gegeben und auch versucht, den Blick für die wahren Dimensionen der jeweiligen Erkrankung und dem sich daraus ableitenden Risiko für den Reisenden zu erläutern.

Im Einzelnen wird zunächst zur aktuellen Lage der Verbreitung von Poliomyelitis und den Änderungen in der Impfstoffversorgung eingegangen.

Die 2012 neu aufgetretenen Erkrankungen durch MERS-CoV haben nach anfänglich durchaus bedrohlichen Entwicklungen sich nun auf niedrigem Niveau stabilisiert, trotzdem sind sie in die Differentialdiagnose nach wie vor einzubeziehen.

Weitaus relevanter für den internationalen Tourismus ist die rasche Ausbreitung von Chikungunya-Infektionen vor allem im mittel- und südamerikanischen Raum, eine Erkrankung, die vor allem durch postinfektiöse Arthralgien durchaus zu bedeutenden Beeinträchtigungen führen kann.

Ein rezenter Gelbfieber-Ausbruch in Angola und die Weltlage zu Gelbfieber werden kurz dargestellt, ebenso wie die momentane Situation zur Denguefieber-Epidemiologie.

Noch nicht wirklich in seiner Auswirkung auf den Tourismus abzuschätzen ist ein Ausbruch von Zika-Virus, ein Flavivirus, am amerikanischen Kontinent und hier wiederum vor allem in Brasilien. Mittlerweile kann man als bewiesen annehmen, dass ZIKA Infektionen in der Gravidität ein hohes fetales Missbildungsrisiko zur Folge haben und dass auch Guillain Barré Erkrankungen in der Folge auftreten können.

Arthropoden: Fakten und Mythen. Herausforderungen der Medizinischen Entomologie im 21. Jahrhundert

Horst Aspöck

Institut für Spezifische Prophylaxe und Tropenmedizin, Medizinische Parasitologie, Medizinische Universität Wien, Kinderspitalgasse 15, 1090 Wien, Österreich

horst.aspoeck@meduniwien.ac.at

Es gibt mehr als eine Million beschriebener (valider) Arthropoden-Spezies (Insekten, Milben, Zecken, Spinnen, Skorpione, Tausendfüßer u.a.), die tatsächliche Zahl wird auf über zehn Millionen geschätzt, vermutlich ist sie sogar (wesentlich) höher. Zwar können mehrere tausend Arten – als Erreger oder als Vektoren – mit Krankheiten des Menschen in kausalem Zusammenhang stehen, jedoch sind ca. 99% aller durch Arthropoden bedingten Erkrankungen auf weniger als 300 Arthropoden-Spezies zurückzuführen.

Arthropoden können die Gesundheit des Menschen in vielfacher Weise bedrohen, als Parasiten von Geweben oder Hohlorganen, als Erreger toxischer und allergischer Reaktionen, als Zwischenwirte von Helminthen, als Ursachen psychischer Irritationen – als Vektoren kommt ihnen allerdings der weitaus höchste Stellenwert zu. Die große Bedeutung von Arthropoden als Überträger von Krankheitserregern ist erst erstaunlich spät, im Wesentlichen erst ab dem letzten Drittel des 19. Jahrhunderts, erkannt worden, heute kennen wir Erreger ebenso wie Überträger der weitaus meisten dieser Erkrankungen, wenn auch mit Neuentdeckungen durchaus noch gerechnet werden muss.

Unter den Erregern nehmen – abgesehen von Plasmodien, Trypanosomen und Leishmanien – die Viren, im Wesentlichen die Arboviren, den höchsten Stellenwert ein und sie werden diese Position auch zum Ende des Jahrhunderts und darüber hinaus behalten – auch wenn vermutlich gegen einige weitere Arboviren mit besonders großer Bedeutung Impfstoffe verfügbar sein werden. Hingegen darf man mit zunehmend erfolgreichen Strategien gegen Malaria, Schlafkrankheit, Morbus Chagas und Leishmaniosen rechnen.

Unser Jahrhundert ist durch drei entscheidende unvermeidbare Entwicklungen gekennzeichnet:

- (1) Bevölkerungswachstum (derzeit über 7,4 Milliarden, Ende des Jahrhunderts vermutlich um 11 Milliarden);
- (2) Klimaerwärmung (global um mindestens 2°C, möglicherweise erheblich höher);
- (3) eskalierende Globalisierung und Migrationen in allen Formen (derzeit z.B. 1 Milliarde Reisende/Touristen pro Jahr, 65 Millionen Flüchtlinge, Verfrachtung von Tieren, Pflanzen und riesigen Mengen von Gütern auf dem Land, mit Schiffen und in Flugzeugen über immer größere Entfernungen in immer kürzerer Zeit).

Alle diese Entwicklungen sind mit einer Zunahme von Krankheiten durch Arthropoden verbunden – bedingt durch Einschleppung oder auch natürliche

Ausbreitung von Erregern und Überträgern, durch Ballungsräume in Ländern mit zunehmend wachsender Bevölkerung und viele andere Faktoren.

Viele Arthropoden haben schon in früher Zeit die Menschen zu absurden Vorstellungen verleitet, aber auch in unserer aufgeklärten Zeit gibt es eine ganze Menge unsinniger – abstruser, falscher, irriger – Meinungen, von denen viele im Zusammenhang mit Krankheiten, die durch Arthropoden bedingt sind, stehen. Die Hartnäckigkeit, mit der manch ein Unsinn auch vom intelligenten Menschen vertreten und verbreitet wird, ist zumindest bemerkenswert.

POSTERS

In co-cultures of *Mycoplasma hominis* with *Trichomonas vaginalis* type strain G3, displacement of *M. hominis* strain Mh475 by MhAKH led to loss of mycoplasmal TET-resistance and loss of parasitic stress response

Ursula Fürnkranz^{1,2}, Julia Walochnik¹, Birgit Henrich²

¹ Institute for Specific Prophylaxis and Tropical Medicine; Medical University of Vienna Kinderspitalgasse 15; 1090 Vienna

² Institute of Medical Microbiology and Hospital Hygiene; Heinrich-Heine-University; Universitätsstraße 1; 40225 Düsseldorf

ursula.fuernkranz@meduniwien.ac.at

Mycoplasma hominis is a sexually transmitted bacterium, found in the normal human vaginal flora. Trichomonas vaginalis, the causative agent of trichomonosis, has often been isolated harbouring Mycoplasma hominis. This co-existence has been described to influence the drug susceptibility and cytotoxicity of the parasite; however, consequences for M. hominis have not been investigated so far. The aim of the presented study was to investigate an influence of passaging M. hominis through T. vaginalis concerning the drug susceptibility of the bacterium. Stable cocultures of TvG3 (type strain) and Tv50138 with *M. hominis* isolate 475 (harbouring a *tet*M transposon mediating resistance to tetracycline) have been successfully established. At several time points of co-culturing, M. hominis was recultured in mycoplasma culture medium and the drug susceptibilities to tetracycline were tested. Tetracycline resistance was measured using a minimal inhibitory concentration assay (MIC); the presence of the transposon was screened using a specifically designed qPCR with several sets of primers. After several weeks of co-culture with TvG3 Mh475 we observed a loss of resistance to tetracycline due to loss of the transposon. However, sequencing revealed that this was not due to loss of tetM-transposon, but a displacement by the tetM-negative M. hominis strain MhAKH. This was not observed during the same time period of co-culture with Tv50138. Here Mh475 was not displaced and thus remained resistant to tetracycline and the presence of the transposon was proven by qPCR.

Supported by FWF T 625-B22.

Door-to-door survey: Epilepsy in a rural area in southern Tanzania. Prevalence, semiology and treatment gap of epilepsy in Mahenge, Ulanga

<u>Christian Gatterer^{1,4}</u>, Teresa Pfeiffenberger^{1,4}, Matthias Wiesmayr^{1,4}, Thomas Wagner^{2,4}, Andrea Winkler^{3,4}, Erich Schmutzhard^{1,4}

¹Department of Neurology, Medical University Innsbruck, Austria

²Rheuma-Kinder-Klinik, Garmisch Partenkirchen, Germany

 $^{3}\mbox{Centre}$ for Global Health , Institute of Health and Society , University of Oslo , Norway

⁴Mahenge District Hospital, Tanzania

Purpose: A door-to-door survey was carried out in order to estimate the prevalence of active epilepsy in the area around Mahenge, Tanzania. Another aim of the study was to classify the type of epileptic seizures in this area and to estimate the treatment gap.

Method: In total, 5467 people in eight different villages have been screened with a five-item questionnaire for epilepsy. One positive response lead to a thorough physical examination and an interview with the patient or a close family member. Final diagnosis of epilepsy was established by a medical doctor or a mental health nurse. Epileptic seizure types were classified according to an adjusted classification for resource poor countries.

Results: Eighty-five individuals (52 females and 33 males) were identified to suffer from active epilepsy and the prevalence ratio was estimated to be 15.5/1000 (95 % CI: 12.6/1000 –19.2/1000). One third (28/85) of the patients were diagnosed with primarily generalized seizures, while 23 % (19/85) were suffering from partial seizures with secondary generalization and 7 % (6/65) had partial seizures without generalization. Twelve patients were found to have the head nodding syndrome. Predominantly more women were affected from epilepsy than men (18.1/1000 vs 12.7/1000). The peak prevalence was between 16 and 25 years in females and between 36 and 45 years in men. Eighty-eight percent of the people with epilepsy had seen a health institution at least once before because their disease and the treatment gap in the area around Mahenge was 18 %; only 15 out of 85 patients

treatment gap in the area around Mahenge was 18 %; only 15 out of 85 patients did not receive antiepileptic drugs for the control of their seizures. Education at school was affected and diminished due to seizures in two thirds of the patients.

Discussion: This study provides further evidence that the prevalence of epilepsy might be higher in rural areas in sub-Saharan Africa than in western countries. The treatment gap in the area around Mahenge seems to be smaller than in other parts of Tanzania or in neighbouring countries, leading to the conclusion that the establishment of the Mahenge Epilepsy Clinic might help to reduce the treatment gap. More case-control studies are needed in order to identify the causes that might be explain the high epilepsy prevalence in low- and middleincome countries.

Comparison of methods for the quantitative analysis of Strongyle eggs in horse faeces

Barbara Hinney^{1*}, Tim Kaphengst¹, Alexander Tichy², Anja Joachim¹

¹ Institute of Parasitology, Department for Pathobiology, Veterinary University of Vienna, 1210 Vienna, Austria

² Bioinformatics and Biostatistics Platform, Department of Biomedical Sciences, Veterinary University of Vienna, 1210 Vienna, Austria

*barbara.hinney@vetmeduni.ac.at

In modern equine parasitology assays to detect high strongylid egg-shedders and examinations to detect anthelmintic resistance of strongyles in the field by the faecal egg count reduction test (FECRT) are frequently applied [1,2]. For both tests an accurate detection of eggs per gram of faeces (EpG) is necessary. The McMaster method (MC) has long time be considered as gold standard for quantification of strongyle eggs in faeces however high variance and low accuracy especially for low egg counts have been observed. Recently Flotac[®] has been developed as a new quantification method with higher accuracy and less variance [3]. However it is comparatively time consuming. Mini-Flotac[®] (MF) is a simplified version of Flotac[®] [4]. We wanted to compare MC and MF for accuracy and sensitivity in quantification of strongyle eggs in equine faeces as well as their time requirements. 12 equine faecal samples composed of four samples each with low, medium and high egg counts were examined by MF and MC with an analytic sensitivity of 5 and 20 EpG respectively. Each sample was portioned into 24 subsamples of 5 g which were randomly assigned to the different techniques.

Correlation coefficient (r), arithmetic mean, coefficient of variation (CV) as well as the time requirement were measured and calculated for each method. Both methods had a high agreement with a correlation coefficient of r >0.90. Looking at the mean egg count in samples with low egg counts both methods had nearly similar counts. CV was highest in MC. At medium egg counts MF had slightly higher egg counts (EpG=45) than MC (EpG=37) and the lowest CV. In samples with high egg load MC had the lowest (EpG=543) and MF the highest counts (EpG=601) while the CV was highest in MF.

When CV is taken as a measurement for accuracy of the test MF was the most accurate test for low and medium egg counts while MC was the most accurate test for high egg counts. No significant differences of time requirements could be calculated. As both methods showed a high correlation and as CV was not consistently lower in one of the tests changing the method from MC to MF is not considered to increase the validity of tests for FECRT or the identification of high egg shedders.

- 1. von Samson-Himmelstjerna, Pfister, Nielsen. Selective therapy in equine parasite control--application and limitations. Vet Parasitol. 2014 202:95-103
- 2. Coles, Jackson, Pomroy, Prichard, von Samson-Himmelstjerna, Silvestre, Taylor, Vercruysse. The detection of anthelmintic resistance in nematodes of veterinary importance. Vet Parasitol. 2006 136:167-85.

Nodding Syndrome in Mahenge, Tanzania, a 10-year follow up.

<u>Pfeiffenberger T⁴</u>, Wagner T^{1,2}, Wiesmayr M⁴, Gatterer C⁴, Kegele J³, Matuja W⁵, Winkler, AS⁶, Schmutzhard E⁴

¹ Centre for Pediatric and Adolescent Medicine, Division of Pediatric Neurology and Inherited Metabolic Diseases, University of Heidelberg, Heidelberg, Germany;

²German Center for Infection Research (DZIF), Munich, Bonn, Germany; ³ Department of Neurology and Epileptology, Hertie Institute for Clinical Brain Research, University of Tübingen, Tübingen, Germany;

⁴ Department of Neurology, Muhimbili University of Health and Allied Sciences, Dar es Salaam, Tanzania;

⁵ Department of Neurology, Medical University Innsbruck, Innsbruck, Austria ⁶ Department of Neurology, Technische Universität München (TUM), Munich, Germany

Describing the course of Nodding Syndrome and the fate of affected individuals in Mahenge, Tanzania.

Nodding syndrome (NS) is still a poorly understood disorder defined by repetitive nodding of the head (head nodding (HN)) combined with cognitive- or behavioral problems, additional seizures, impaired growth or development. Within the last decade several case series, case-control-surveys and community-based prevalence studies were conducted in South Sudan, northern Uganda and southern Tanzania. Nevertheless, the course of the disorder remains controversial and long-term data are scarce. We intend to report the clinical development of previously reported patients with NS from Mahenge, Tanzania as well as the course of NS.

12/2014 to 4/2015 a study on course and phenotype of NS and epilepsy was performed in Mahenge, Tanzania. Thereby participants from studies conducted in 2005 and 2009 were retrieved for reassessment using a standardized questionnaire and examination schedule including video EEG and cognitive evaluation.

Of the initially reported 62 patients, two had moved and were well according to the families, 5 passed away, 4 of them related to epileptic seizures. 17 patients were lost or failed to re-attend, the rest was re-located and evaluated in detail. Among those 38 patients, 14 (37%) reported persistent HN seizures daily to once a year. HN had stopped in 24 (63%) of the patients of whom 22 remained on antiepileptic treatment. 26 people reported additional, mainly generalized tonic-clonic, seizures that started 0 to 17 years (mean 3 years) after the onset of HN. Compared to 2009, medication had been adjusted in 27 of 38 patients (71%) due to different reasons. Only 13 of 26 patients with additional seizures (50%) experienced additional seizures within the last 12 months though HN persisted in 4 of them. Compared to 2009, seizure frequency was significantly reduced in patients with remaining additional seizures. Even though the majority dropped out of primary school, 21 of 35 (60%) remained independent and able to ensure their own living. 6 were married and 13 were raising own children. None of the patients reported a decline or lost any previously achieved abilities.

Under appropriate antiepileptic treatment, patient with HN in Mahenge, Tanzania may display a more favourable course than reported elsewhere.

Multiple Intestinal Protozoal Parasites and Filariasis in a Migrant from Bangladesh

Trattner Elisabeth, Haas Bernhard

Department of Gastroenterology with Infectious Diseases, LKH Graz Süd-West, Standort West, Graz

Diagnosing fever of unknown origin (FUO) in Austrian long-term residents with a migrant background can be a challenge in daily routine in Austrian public hospitals.

We report a case of a 35 year-old South Asian female diagnosed with multiple parasites. Different methods like enzyme-linked immunosorbent assay (ELISA), western blot and indirect haemagglutination assay (IHA) were used as serological tests. Diagnostic procedures of stool samples included Microscopy and antigen testing against parasites like Cryptosporidium parvum, Giardia lamblia and Entamoeba histolytica. Diagnostic procedures were performed in different Microbiology laboratories in Graz and Vienna and the Department of Medical Parasitology, Institute of Specific Prophylaxis and Tropical Medicine, Medical University of Vienna.

In June 2016 a 35 year old South Asian female from Bangladesh was admitted to our outpatient's clinic for Infectious Diseases and Tropical Medicine complaining of recurrent episodes of fever, abdominal pain, aching limbs, malaise and respiratory symptoms with temporary coughing since several years. The last trip to Bangladesh was in 2015. The past medical history revealed two hospitalizations at a second hospital in Styria, Internal Medicine in 2006 and 2014 for diagnostic

exploration of the previously mentioned symptoms but showed no pathological correlate so far. In our investigations concerning inflammation markers a minimally elevated c-reactive protein of 1,4 mg/dl (ULN 0,5 mg/dl) was detected. The eosinophil count was normal. A CT scan of the chest showed lymphadenopathy of the axilla and abdominal sonography was without any pathological findings.

HIV-1/2 antibody test was negative. Microscopic examination of the stool showed Entamoeba histolytica and eggs of Trichuris trichiura. The serological findings revealed repeatedly raised antibodies against ascaris suum by western blot and highly raised antifilarial antibody titres by ELISA – but latter only once. To rule out pulmonary tuberculosis following a positive result of the Interferon- Gamma Release Assay (IGRA) a bronchoskopy with bronchoalveolar lavage (BAL) was performed.

In suspicion of Filariasis the sampling of BAL was microscopically investigated for helminths but they could not be confirmed in this sample. Meanwhile we started therapy of amoebiasis with metronidazole 500 mg b.i.d for 10 days. Mebendazole 200 mg was administered for Ascaris suum and Trichuris trichiura. Surprisingly when monitoring the therapeutic effect on the soil transmitted nematodes and Entamoeba a stool specimen was microscopically investigated and there another parasite - Blastocystis hominis - was detected.

With regard to Filariasis, the diagnostic results were not satisfying. The serum level of total IgE was not yet investigated. The direct proof of microfilaria molecular diagnostic methods (e.g. Polymerase Chain Reaction) in peripheral blood collected at evening or night time was also not performed yet. Lung biopsy may be required to rule out tropical pulmonary eosinophilia - an immune hyperresponse syndrome caused by microfilaria resulting in progressive pulmonary damage if not

treated [1]. So further investigations will be necessary regarding Filariasis. In the meantime diagnostic procedures regarding persistence of Blastocystis hominis including stool microscopy will be performed at the next outpatient visit.

References: 1. Jai B. Mullerpattan, Zarir F.Udwadia, and Farokh E. Udwadia, Tropical pulmonary eosinaphila-A review, IJMR,2013, 138(3); 295-302

Comparison of tick-screening methods for pathogen detection and impact on results

Anna-Margarita Schötta*, Michiel Wijnveld, Hannes Stockinger, Gerold Stanek

Institute for Hygiene and Applied Immunology, Center for Pathophysiology, Infectiology and Immunology, Medical University of Vienna, Austria, 1090

*anna-margarita.schoetta@meduniwien.ac.at

Ticks can transmit a variety of microorganisms that can lead to human disease. In a previous study 554 ticks have been screened for a number of pathogens including spirochetes of the *Borrelia burgdorferi* sensu lato (*Borrelia*) complex and *Rickettsia* spp.

In the course of a screening for these pathogens two different detection approaches based on the reverse line blot (RLB) hybridization were applied for each of the two groups of bacteria. The RLB consists of two major steps: firstly, a PCR with a biotinylated primer is performed, resulting in biotinylated amplicons. Secondly, those amplicons are hybridized to a membrane containing genus and species specific oligonucleotide probes. A successful hybridization is then visualized by chemiluminescence.

For the detection of *Borrelia* the methods differed simply in the primer pair used for PCR. As both primer pairs target the 5S-23S intergenic spacer (IGS) region the probes for detection by RLB were identical. A sensitivity testing showed the same detection limit for both PCRs. However, when applied to tick DNA samples a significant difference was observed. The overall prevalence obtained with the different primer pairs was 24.7% and 13.5% with primer pair 1 and 2, respectively. Species distribution within the *Borrelia* positive ticks was similar with both methods, except for *B. valaisiana* which was less often detected by primer pair 2 (16.0% vs. 26.3%), and for *B. spielmanii* which was only detected when primer pair 1 was used.

Detection of *Rickettsia* spp. was achieved by performing two completely independent RLBs based on different targets. One target gene is based on the 16S rRNA gene while the more recently designed RLB is based on the 23S-5S IGS of *Rickettsia* spp. The overall prevalence detected was 15.5% and 13.7% with RLB1 (16S) and RLB2 (23S-5S IGS), respectively. By the use of RLB1 the species *R. raoultii* was more often detected when compared to RLB2 (44.2% vs. 35.5%) while *R. slovaca* could only be detected by RLB2.

Our results show that similar methods, even if they have the same sensitivity in the evaluation process, can lead to a completely different prevalence of genera and species detected. Hence, detection methods should be chosen with special consideration.

Sponsored by:

