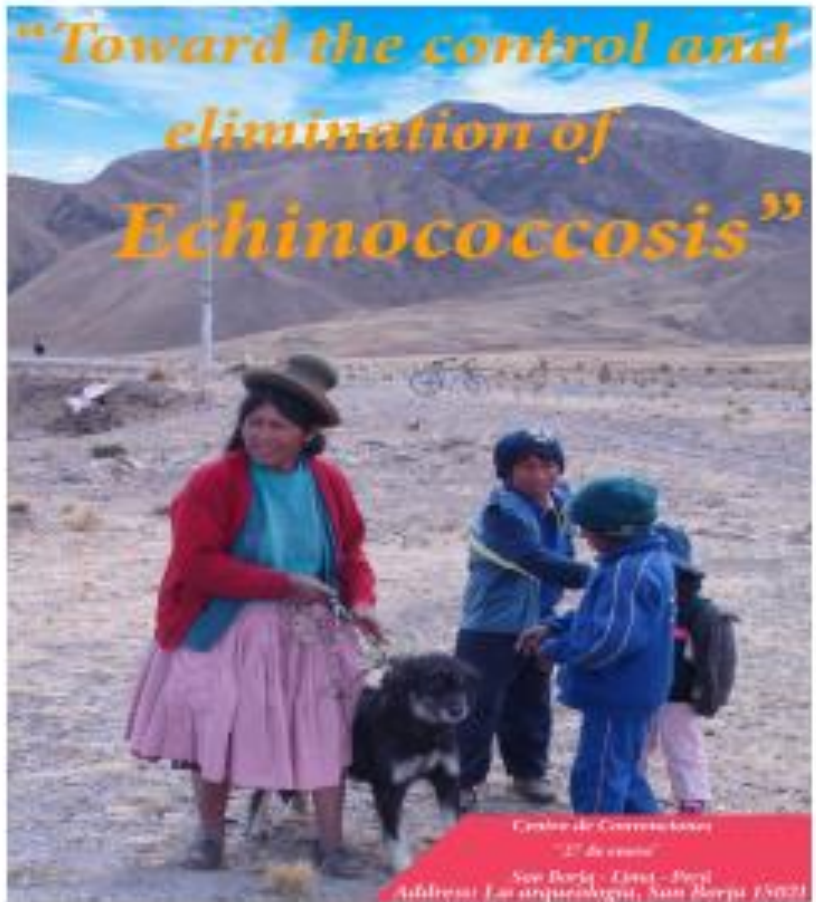




# ABSTRACT BOOK

WORLD CONGRESS OF ECHINOCOCCOSIS  
AND 2ND PERUVIAN CONGRESS  
OF ECHINOCOCCOSIS

October  
29-31  
Lima, Peru



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## WELCOME LETTER

On behalf of the organizers, we are pleased and honored to announce and welcome all of you to the XXVIII World Congress of Echinococcosis, which will be held in Lima, Peru, from October 29-31, 2019. The Congress is organized under the sponsorship of the The World Association of Echinococcosis (WAE), the Peruvian Association of Hydatidology (PAH), Universidad Nacional Mayor de San Marcos and Universidad Peruana Cayetano Heredia and the Ministry of Health. Despite the enormous efforts to control and eliminate Echinococcosis in the world, many countries from Latin America, Asia, Africa and Europe still report high prevalence and incidence rates of Cystic Echinococcosis (CE, *E. granulosus*) in humans and animals. Furthermore, there are two other important Echinococcosis; first, Alveolar Echinococcosis (AE, *E. multilocularis*) that represents a serious disease in several countries in Asia, and second, Neotropical Echinococcosis (NE, *E. vogeli*) which affect a considerable region in Latin America.

Country-based Control Programs have been struggling these parasites for decades with different successes or failures. New and recent attempts have been implemented in other countries, and their impact on Echinococcus elimination would come in the near future. We expect that this XXVIII World Congress of Echinococcosis would be a remarkable scientific event that will bring the interest of all distinguish experts and professionals in the field from all over the World. This meeting will create the environment to exchange experiences, research, outstanding findings, collaboration, and enhance the relationships among all participants.

The Congress is addressing to physicians, veterinarians, biologists, parasitologists, pharmacologists, infectious diseases specialists, surgeons, epidemiologists, public health practitioners, laboratory specialists, economists, and all who are involved and/or interested in Echinococcosis. The Congress will be carried out on round tables, scientific sessions, symposium, and poster sessions. The official language of the Congress is English.

We thank all of you in advance for your participation and commitment to defeat this zoonotic parasite in the World. Your presence in this World Congress will give the prestige of the outstanding scientific event.

Welcome to Lima, we hope you enjoy your stay in our beautiful country.

ORGANIZING COMMITTEE

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## ORGANIZING COMMITTEES

### LOCAL ORGANIZING COMMITTEE

Dr. César Náquira (President)

PhD. Saúl Santivañez (Treasurer)

PhD. César Gavidia (Secretariat)

### CO-ORGANIZERS

Dr. Karim Pardo (Congress Venue)

### SCIENTIFIC COMMITTEE

Dr. Edmundo Larrieu (Vice President of World Association of Echinococcosis, Argentina)

Dr. Marshall Lightowlers (University of Melbourne, Australia)

Dr. Donald McManus (QIMR Berghofer Medical Research Institute)

Dr. Maria Mar Siles (Instituto de Recursos Naturales y Agrobiología de Salamanca, Spain)

Dr. Enrico Brunetti (University of Pavia, Italy)

Dr. Okan Akhan (Hacettepe University, Turkey)

Dr. Thomas Junghanss (University of Heilderberg, Germany)

Dr. Philip Craig (University of Salford, United Kingdom)

Dr. Peter Deplazes (University of Zurich, Switzerland)

Dr. Katherina Vizcaychipi (Instituto Nacional de Enfermedades Infecciosas - ANLIS "Dr. Carlos G. Malbran", Argentina)

Dr. Antonio Meneses da Silva (Past President of the World Association of Echinococcosis)

Dr. Ciro Ferreira (President of the World Association of Echinococcosis)

Dr. Nazmiye Altintas (Ege University Faculty of Medicine, Turkey)

Dr. Hugo Garcia (Universidad Peruana Cayetano Heredia, Peru)

Dr. Armando Gonzalez (Universidad Nacional Mayor de San Marcos, Peru)

Dr. Manuela Verastegui (Universidad Peruana Cayetano Heredia, Peru)

## GENERAL INFORMATION

### HEALTH PRECAUTIONS

Yellow fever vaccination is not required to enter our country unless you are traveling from a region where yellow fever is endemic.

Tap water is usually not safe to drink in Peru, we suggest to purchase bottled water.

Time: GMT -5

Peruvian currency is the Sol (S/), exchange rate is about of 1 US \$ = S/. 3.38

### PERU\*

Peru is located in the western part of South America. Its territory borders Ecuador, Colombia, Brazil, Bolivia and Chile. It covers 1,285,215 km<sup>2</sup> of land and 200 nautical miles of the Pacific Ocean, as well as 60 million hectares in Antarctica. It has an estimated population of over 31.5 million inhabitants.

It is the third largest country in South America and one of the 20 largest in the world - its surface area covers more than the territories of Spain and France combined. Being a consultative party to the Antarctic Treaty, it has a scientific station called 'Machu Picchu' on this continent.

The country is divided into 25 regions or departments, including the constitutional province of El Callao, where Peru's main port is located. The capital is Lima, a city founded in the year 1535 by the Spanish, and becoming the capital of the Viceroyalty of Peru. After independence in 1821, it became the capital of the republic.

Peru is a highly diverse country. It has 11 ecoregions and 84 of the 117 life zones that exist in the world. It has an enormous range of landscapes due to its geographical conditions, which in turn give it a great diversity of natural resources. Three main regions can be identified in its territory, which is the traditional way of dividing it according to altitudes: Coast, Mountains and Jungle.

### LIMA\*

'Lima is Peru', as the celebrated Peruvian writer Abraham Valdelomar quite rightly said. You can encounter all our races and customs in its colonial streets and modern districts that look out onto the Pacific Ocean, a synthesis of our cultural diversity and our incomparable gastronomy, as well as certain manifestations that allow us to understand the complex history of our country a little better.

But beyond this historical-cultural aspect of Peru's capital, Lima is also film and theater, good music, shopping and art galleries, nights

out, museums and a different cityscape that looks out onto the sea, and in which the present and the past combine to create a city like few in the world.

Handicrafts, alpaca, jewelry, silverware, painting, photography, Lima is the best place to go shopping and it has an interesting range of shopping centers, art galleries, workshops and craft shops and ateliers where you can buy the best Peruvian products from every region.

Its range of restaurants, bars and markets has made it recognized as the gastronomic capital of the region. The city houses three of the 50 best restaurants in the world and nine of the 50 best in Latin America, according to the 2016 list compiled by Restaurant magazine. But it also has an important pre-Hispanic legacy, with archaeological sites in the middle of the city, such as the huacas of Mateo Salado, Pucllana or Huallamarca, and sanctuaries such as Pachacamac, located on the southern outskirts of the city.

### VENUE

Lima Convention Center "27 DE ENERO",

Address: Calle La Arqueología, San Borja 15021

Language: The official language of the Congress is English

Dates: 28-31 October 2019

Registration and Information: Desk Opened desk on the following dates:

October 28<sup>th</sup> Pre congress course 09:00-18:00

Welcome and Opening ceremony  
18:30 - 22:00

October 29<sup>th</sup>-31<sup>st</sup>: 09:00 - 19:00

After the registration you will receive your badge and Congress kit. You are kindly invited to wear your badge during the Congress sessions and social events.

### SESSIONS

You are kindly invited to be on session halls on time, as all the sessions will begin as per schedule.

### POSTER PRESENTATIONS

A different set of posters will be on display from October 29<sup>th</sup> to 31<sup>st</sup>. Posters will be displayed as per the posters number indicated in the Poster Program in Huantile 2 room. Poster presenters are requested to stand by their posters during the breaks.

\*Source: Tourist information <https://www.peru.travel>



## CONGRESS PROGRAM

### Monday, October 28

Time	Activity (Sala Mangamarca)
08.30 - 18.00	<b>Registration</b>
09.00 – 18:00	Pre- Congress Course: <b>Advances in Diagnosis and Management of Echinococcosis.</b>
18:30 – 19.30	Welcome and Opening Ceremony Ministry of Health Ministry of Agriculture – Peruvian Veterinary Service (SENASA) Universidad Nacional Mayor de San Marcos Universidad Peruana Cayetano Heredia World Association of Echinococcosis (Dr. Ciro Ferreira)
20.00 - 22.00	Social Confraternity

### Tuesday, October 29

Time	Sala Mangamarca	Sala Huantile 2	Sala Huantile 1
Oct. 29 09:00 – 11:00	<b>Round table 1</b> Cystic Echinococcosis in Peru: Current Situation  <u>CHAIR</u> César Náquira, Dr (Peru)  <u>INVITED SPEAKERS</u> - Saúl Santivañez, PhD (Peru) <i>“Cystic echinococcosis in Peru: Current projects in CGH”</i>		Poster session 1 (Posters from 1 – 40)

	<ul style="list-style-type: none"> <li>- César Gavidia, PhD (Peru) “Contributing to control Cystic Echinococcosis”</li> <li>- Ubaldo Flores, Mg (Peru) Peruvian Veterinary Service (SENASA) “The Control and Prevention of Hydatidosis / Cystic Echinococcosis Pilot Program in Peru”</li> <li>- José Bustamante, MV (Peru) Peruvian Ministry of Health “Cystic Echinococcosis in Peru”</li> </ul>		
11:00 – 11:30	<b>Coffee break</b>		
Oct. 29 11:30 – 13:00	<p><b>Symposium 1:</b> Imaging methods in the diagnosis and follow up of human Cystic Echinococcosis</p> <p><u>CHAIR</u> Francesca Tamarozzi, PhD (Italy)</p> <p><u>INVITED SPEAKERS</u></p> <ul style="list-style-type: none"> <li>- Thomas Junghanss, MD (Germany) “US, CT and MRI in cystic echinococcosis”</li> <li>- Marija Stojkovic, MD (Germany) “US in follow-up (watch and wait) overview of evidences gathered so far”</li> </ul>	<p><b>Symposium 2:</b> Molecular biology of the species of <i>Echinococcus</i></p> <p><u>CHAIR</u> Dominique Vuitton, PhD (France)</p> <p><u>INVITED SPEKAER</u></p> <ul style="list-style-type: none"> <li>- Thomas Romig, PhD (Germany) “Species and genotypes: what is new?”</li> </ul> <p><u>SCIENTIFIC ORAL PRESENTATIONS</u></p> <p>1. Nazmiye Altintas, MD (Turkey)</p>	<p>Poster session 1 (Posters from 1 – 40)</p>

	<p>- Francesca Tamarozzi, PhD  <i>“Update on the WHO-IWGE work on imaging”</i></p>	<p><i>“Distribution map of Echinococcus granulosus genotypes in Turkey”</i></p> <p>2. Jenny Knapp (France)  <i>“European Alveolar echinococcosis genotyping: an EmsB microsatellite analysis”</i></p> <p>3. Gérald Umhang (France)  <i>“Probability of detection of Echinococcus multilocularis DNA in fox fecal samples”</i></p>	
13:00 – 14:30	<b>Lunch</b>		
Oct. 29 14:30 – 16:00	<p><b>Symposium 3:</b> Immunological methods of the diagnosis and follow up of human Cystic Echinococcosis</p> <p><u>CHAIR</u>  Mar Siles, PhD (Spain)</p> <p><u>INVITED SPEAKERS</u></p> <p>- Manuela Verastegui, PhD (Peru)  <i>“Challenges to improve Serologic Diagnosis of Cystic Echinococcosis”</i></p> <p><u>SCIENTIFIC ORAL PRESENTATIONS</u></p> <p>4. Francesca Tamarozzi (Italy)  <i>“Diagnostic performances of commercial elisa, iha, and wb for the diagnosis of echinococcal vs non-echinococcal lesions:</i></p>	<p><b>Symposium 4:</b> Diagnosis and treatment / prevention of cystic echinococcosis (intermediate hosts, animal models)</p> <p><u>CHAIR</u>  Cesar Gavidia, PhD (Peru)</p> <p><u>INVITED SPEAKERS</u></p> <p>- Celina Elissondo, Dr. (Argentina)  <i>“Can Pharmacochemical Strategies be useful to optimize the Anti-Echinococcal Efficacy of Albendazole?”</i></p> <p>- David Heath, PhD (New Zealand)  <i>“EG95 vaccine testing in China. Factory production, safety and efficacy</i></p>	<p>Poster session 1  (Posters from 1 – 40)</p>

	<p><i>a retrospective analysis of data of a single reference centre</i></p> <p>5. Bao Haihua (China) <i>“Serum Ferritin combined with MRI in identifying IgG negative Application study of hepatic cystic echinococcosis and hepatic cysts”</i></p> <p>6. Laurence Millon (France) <i>“Value of soluble programmed death-1 (sPD-1) and sPD-ligand-1 (sPDL-1) as immunological biomarkers for the follow-up of cystic echinococcosis in Tunisian pediatric patients”</i></p>	<p><i>to Registration and to large scale production”</i></p> <p><b><u>SCIENTIFIC ORAL PRESENTATIONS</u></b></p> <p>7. Jun Li (China) <i>“A platform for identifying novel carbazole aminoalcohols against echinococcosis”</i></p> <p>8. Ariel Naidich (Argentina) <i>“Towards an Experimental Model For Studying Cyst Formation”</i></p> <p>9. Katherina Vizcaychipi (Argentina) <i>“Serodiagnostic Study of Cystic Echinococcosis in Sheep from areas of Amazonian Dominion, Province of Misiones, Argentina”</i></p>	
16:00 – 16:30	<b>Coffee break</b>		
Oct. 29 16:30 – 17:30	<p><b>Conference 1:</b> Socioeconomic impact of Echinococcosis in endemic countries</p> <p><b><u>CHAIR</u></b> Paul Torgerson, PhD (Switzerland)</p> <p><b><u>INVITED SPEAKER</u></b> Christine Budke, PhD (USA)</p>		<p>Poster session 1 (Posters from 1 – 40)</p>

	<i>“Overview of the Socioeconomic Impact of Echinococcosis in Endemic Countries”</i>		
17:30 – 19:00	World Association of Echinococcosis - General Assembly (Sala Mangomarca)		

### Wednesday, October 30

Time	Sala Mangomarca	Sala Huantile 2	Sala Huantile 1
Oct. 30 09:00 – 11:00	<p><b>Round table 2:</b> Situation of Cystic Echinococcosis around the world</p> <p><u>CHAIR</u> Edmundo Larrieu, PhD (Argentina)</p> <p><u>INVITED SPEAKER</u></p> <ul style="list-style-type: none"> <li>- Adriano Casulli, PhD (Italy) <i>“The burden of cystic and alveolar echinococcosis in Europe”</i></li> <li>- David Heath, PhD (New Zealand) <i>“National Control Program of Cystic Echinococcosis in New Zealand. Cystic Echinococcosis in Australia. Cystic Echinococcosis in China”</i></li> <li>- Julio Sayes, MSc (Uruguay)</li> </ul>		Poster session 2 (Posters from 41 – 62)

	<i>“National Control Program of Cystic Echinococcosis in Uruguay: achievements and future challenges”</i>		
11:00 – 11:30	<b>Coffee break</b>		
Oct. 30 11:30 – 13:00	<p><b>Symposium 5:</b> Surgery and other intervention options for Cystic Echinococcosis treatment in humans</p> <p><u>CHAIR</u> Okan Akhan, MD, Prof. (Turkey)</p> <p><u>INVITED SPEAKERS</u></p> <ul style="list-style-type: none"> <li>- Enrico Brunetti, MD (Italy) <i>“PAIR technique: Overview”</i></li> <li>- Ciro Ferreira, MD (Uruguay) <i>“Tratamiento adventicial selectivo y Desconexión Quisto Biliar en Q.H.H”</i></li> <li>- Okan Akhan, MD, Prof. (Turkey) <i>“Catheterization and MoCaT techniques: What is new?”</i></li> </ul> <p><u>SCIENTIFIC ORAL PRESENTATIONS</u></p> <p>10. Karima Achour (Algeria) <i>“Algerian experience in medico-surgical management of hydatid disease since the</i></p>	<p><b>Symposium 6:</b> Diagnosis and classification of human Alveolar Echinococcosis</p> <p><u>CHAIR</u> Peter Deplazes, Professor (Switzerland)</p> <p><u>INVITED SPEAKERS</u></p> <ul style="list-style-type: none"> <li>- Beat Müllhaupt, Prof. (Switzerland) <i>“Clinical and laboratory diagnosis of Alveolar Echinococcosis”</i></li> <li>- Tilmann Graeter (Germany) <i>“Imaging classification of Alveolar Echinococcosis”</i></li> </ul> <p><u>SCIENTIFIC ORAL PRESENTATIONS</u></p> <p>11. Wenya Liu (China) <i>“Evaluation of Remnant Liver Function in Patients with Hepatic Alveolar Echinococcosis (HAE) with Energy Spectrum CT”</i></p> <p>12. Dominique Vuitton (France)</p>	<p>Poster session 2 (Posters from 41 – 62)</p>

	<i>World Congress of WAE in Algeria in 2017</i>	<p><i>“Ae Hepatic Lesions : Is There Any Correlation Between Microcalcifications And Fdg Pet-Ct Metabolic Activity ?”</i></p> <p>13. Bao Haihua et al. (China) <i>“Gd-EOB-DTPA-enhanced MRI to evaluate the lesion and liver function in Hepatic Alveolar Echinococcosis”</i></p> <p>14. Philipp Kronenberg (Switzerland) <i>“Early serological diagnosis of human alveolar echinococcosis”</i></p>	
13:00 – 14:30	<b>Lunch</b>		
Oct. 30 14:30 – 16:00	<p><b>Symposium 7:</b> Anti-parasitic drug treatment of Human Cystic Echinococcosis</p> <p><u>CHAIR</u> Thomas Junghanss, MD (Germany)</p> <p><u>INVITED SPEAKERS</u></p> <ul style="list-style-type: none"> <li>- Marija Stojkovic, MD (Germany) <i>“Albendazole Treatment of Cystic Echinococcosis evidence of Cyst Stage Specific Efficacy”</i></li> <li>- Leonardo Uchiumi, MD (Argentina)</li> </ul>	<p><b>Symposium 8:</b> Treatment of Alveolar Echinococcosis</p> <p><u>CHAIR</u> Beate Grüner, PD Dr.med (Germany)</p> <p><u>INVITED SPEAKERS</u></p> <ul style="list-style-type: none"> <li>- Beat Müllhaupt, Professor (Switzerland) <i>“Is it possible to stop benzimidazole treatment in patients with inoperable alveolar echinococcosis?”</i></li> <li>- Beate Grüner, PD Dr.med (Germany)</li> </ul>	<p>Poster session 2 (Posters from 41 – 62)</p>

	<p><i>"Global access to Benzimidazoles: preliminary results of an ongoing survey"</i></p> <p>- Marija Stojkovic MD (Germany), Leonardo Uchiumi MD (Argentina), Thomas Junghanss MD (Germany) <i>Interactive case series: "Would you treat this patient with albendazole?"</i></p> <p><u>SCIENTIFIC ORAL PRESENTATIONS</u></p> <p>15. Adriano Casulli (Italy) <i>"Patent on Salts having Benzimidazolic Compounds: Focus on Sodium Salt of Ricobendazole (Na-Rbz) and its Enantiomers"</i></p>	<p><i>"AE at University Hospital of Ulm (UKU). Diagnosis, treatment and follow up data from 280 AE- patients first presentation from 01.01.2011-31.12.2018"</i></p> <p><u>SCIENTIFIC ORAL PRESENTATIONS</u></p> <p>16. Yingmei Shao (China) <i>"East meets West in pushing the envelope of End-stage Alveolar Echinococcosis Surgery: Ex Vivo Liver Resection and Autotransplantation"</i></p> <p>17. Dominique Vuitton (France) <i>"Is radical surgery still the gold standard for European patients with alveolar echinococcosis?"</i></p> <p>18. Yi Jiang (China) <i>"To predict the correlation between calcification and its biological activity of Hepatic alveolar echinococcosis"</i></p> <p>19. Renyong Lin (China) <i>"Immune exhaustion of T cells in alveolar echinococcosis patients and its reversal by blocking checkpoint receptor TIGIT in a murine model"</i></p>	
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16:00 – 16:30		Coffee break	
Oct. 30 16:30 – 18:00	<p><b>Round table 3:</b> Countries with control / elimination programs of Cystic Echinococcosis</p> <p><u>CHAIR</u> Pedro Moro, MD, MPH (CDC, USA)</p> <p><u>INVITED SPEAKERS</u></p> <ul style="list-style-type: none"> <li>- Nazmiye Altintas, PhD (Turkey) <i>“Control / Elimination Programme Of Cystic Echinococcosis In Turkey”</i></li> <li>- Marshall Lightowlers, PhD (Australia) <i>“Key considerations on implementation, monitoring, evaluation of echinococcosis control actions”</i></li> <li>- Paul Torgerson, PhD (Switzerland) <i>“Control programme Kyrgyzstan”</i></li> <li>- Ran Zhiguang (China) <i>“Large-scale Vaccination of EG95 Vaccine Significantly Interfered with the Dog-sheep/goat Transmission Chain of Hydatidosis”</i></li> </ul>		Poster session 2 (Posters from 41 – 62)

<p>Oct. 30 18:00 – 19:30</p>	<p><b>Scientific Session 1 (oral presentations)</b></p> <p><u>CHAIR:</u> Malika Kachani, PhD (USA)</p> <p>20. Victor del Rio (UK) <i>“Records of deaths attributed to Echinococcosis in Brazil: 1995-2016”</i></p> <p>21. Joanne Widdicombe (UK) <i>“Mathematical modelling of Echinococcus granulosus to identify optimal intervention strategies: a case study from five pilot interventions in the Peruvian highlands”</i></p> <p>22. Nelly Huamani (Peru) <i>“Epidemiological Surveillance of Human Hydatidosis between the years 2018 at the beginning of the year 2019 in the Ayacucho’s Region – Peru”</i></p> <p>23. Hao Wen (China) <i>“Establishment and Innovative Practice of Integration System of Prevention, Diagnosis &amp; Management (ISPDM) for Human Echinococcosis”</i></p> <p>24. Daphne León (Peru) <i>“Educational tools for human cystic echinococcosis prevention among school students on an endemic area of Peru”</i></p>	<p><b>Scientific Session 2 (oral presentations)</b></p> <p><u>CHAIR:</u> Dominique Vuitton, PhD (France)</p> <p>26. Jianhua Wang (China) <i>“In vitro and in vivo efficacy of DNA damage repair inhibitor Veliparib combined with artesunate against Echinococcus granulosus”</i></p> <p>27. Jianhua Wang (China) <i>“Effect of siRNA interference targeting EgRPS9 gene on DNA oxidative damage of Echinococcus granulosus protoscoleces”</i></p> <p>28. Christian Hidalgo (Chile) <i>“Multiple haplotypes of Echinococcus granulosus sensu stricto in single naturally infected intermediate hosts”</i></p> <p>29. Gérald Umhang (France) <i>“Presence of E. multilocularis in Eastern Europe: EmsB microsatellite analyses revealed exclusive influence of European historical focus”</i></p>	<p><b>Internal meeting PERITAS / Initiative to Control CE</b></p>
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	25. Wang Qian (China) <i>“Effectiveness of ten years of control of E.multilocularis by deworming owned dogs using praziquantel in a highly endemic area of Tibetan communities, China”</i>		
20:00	<b>Congress Dinner</b> <b>World Association of Echinococcosis (Dr. Ciro Ferreira) – Awards and official recognitions)</b>		

### Thursday, October 31

Time	Sala Mangomarca	Sala Huantile 2	Sala Huantile 1
Oct. 31 09:00-11:00	<p><b>Round table 4:</b> Continental project experiences on Cystic Echinococcosis</p> <p><u>CHAIR</u> Adriano Casulli, PhD (Italy)</p> <p><u>INVITED SPEAKERS</u></p> <ul style="list-style-type: none"> <li>- CESSARi Thomas Romig, PhD (Germany) <i>“CESSARi: Research on Cystic Echinococcosis in Sub-Saharan Africa”</i></li> <li>- HERACLES</li> </ul>		<p>Poster session 3 (Posters from 63 – 91)</p>

	<p>Adriano Casulli, PhD (Italy)  <i>“The international impact of HERACLES collaborative project on cystic echinococcosis”</i></p> <p>- ERCE  Francesca Tamarozzi, PhD (Italy)  <i>“The state of European Register of Cystic Echinococcosis (ERCE) 5 years after its inception”</i></p> <p>- South American Initiative  Edmundo Larrieu, PhD (Argentina)  <i>“The South American Initiative for the surveillance, control and prevention of Cystic Echinococcosis/Hidatidosis”</i></p> <p>- PERITAS  Adriano Casulli, PhD (Italy)  <i>“Molecular-epidemiological studies on pathways of transmission and long lasting capacity building to prevent cystic echinococcosis infection: outline of PERITAS project”</i></p> <p>- PERITAS  Mar Siles, PhD (Spain)  <i>“Wp2_Molecular/Serological Epidemiological Studies Recombinant Antigens And Serological Tests”</i></p>		
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	<ul style="list-style-type: none"> <li>- NDTND Adriano Casulli, PhD (Italy) <i>“ERANET-LAC project: Development of new diagnostic and treatment options for helminthic neglected diseases (NDTND)”</i></li> </ul> <p><b><u>SCIENTIFIC PRESENTATIONS</u></b></p> <ul style="list-style-type: none"> <li>- Leonardo Uchiumi (Argentina) <i>“Ultrasound screenings in Rio Negro province, Argentina: preliminary results from stage 1 of PERITAS project”</i></li> <li>- Gerardo Acosta (Chile) <i>“Ultrasound screenings in Limarí province, Chile: preliminary results from stage 1 of PERITAS project”</i></li> <li>- Saul Santivañez (Peru) <i>“Ultrasound screenings in Communities of Junin, Peru: Preliminary results from stage 1 of PERITAS project”</i></li> </ul>		
11:00 – 11:30	<b>Coffee break</b>		
Oct. 31  11:30 – 13:00	<p><b>Symposium 9: WHO-IWGE</b></p> <p><b><u>CHAIRS</u></b> Thomas Junghanss, MD (Germany) Okan Akhan, MD, Prof. (Turkey)</p>	<p><b>Symposium 10: EcoHealth / OneHealth</b></p> <p><b><u>CHAIR</u></b> Gerardo Acosta, PhD (Chile) <i>“One Health Symposium”</i></p>	<p>Poster session 3 (Posters from 63 – 91)</p>

	<p><u>INVITED SPEAKERS</u></p> <ul style="list-style-type: none"> <li>- Bernadette Abela-Ridder PhD (WHO, Geneva, Switzerland) <i>“Reinserting CE and AE in the New NTD roadmap and Universal Health Coverage”</i></li> <li>- Thomas Junghanss MD (Germany) <i>“The ‘CE Technical Manual’: reaching out to health services and patients”</i></li> <li>- Laurence Millon PhD (France) <i>“Update from WHO-IWGE - AE Clinical group. Building the AE Technical Manual”</i></li> <li>- Edmundo Larrieu PhD (Argentina) <i>“Vigilancia y control de Echinococosis quística”</i></li> <li>- Marshall Lightowlers (Australia) <i>“IWGE subgroup Control of CE”</i></li> <li>- Paul Torgerson PhD (Switzerland) <i>“AE control, an update”</i></li> <li>- Dominique A. Vuitton PhD (France) <i>“International consensus on terminology in the field of echinococcosis”</i></li> </ul>	<p><u>INVITED SPEAKERS</u></p> <ul style="list-style-type: none"> <li>- Malika Kachani, PhD (USA) <i>“One Health: the reality out there” Translating theory into practice?</i></li> <li>- Peter Deplazes, PhD (Switzerland) <i>Echinococcosis in Europe: transmission ecology, diagnosis and control in a Swiss “One Health” network”</i></li> <li>- David Jenkins, PhD (Australia) <i>“One Health and the evolving epidemiology of Echinococcus granulosus in Australia”</i></li> </ul> <p><u>SCIENTIFIC ORAL PRESENTATIONS</u></p> <p>30. Giulia Paternoster (Switzerland) <i>“Spatial disease modelling to identify potential environmental determinants of human echinococcosis in Kyrgyzstan ”</i></p> <p>31. Rudi Cassini (Italy) <i>“A One-Health evaluation of the socio-economic impact of cystic echinococcosis in the Veneto region, a hypoendemic area in Northern Italy”</i></p>	
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	Discussion		
13:00 – 14:30	<b>Lunch</b>		
Oct. 31	<p><b>Symposium 11:</b> Diagnosis of Echinococcosis in definitive host</p> <p><u>CHAIR</u> Graciela Santillán, Dr (Argentina)</p> <p><u>INVITED SPEAKERS</u></p> <ul style="list-style-type: none"> <li>- Cristian Alvarez, MSc (Switzerland) “Update on knowledge of transmission of Echinococcus to humans”</li> <li>- Edmundo Larrieu (Argentina) “Surveillance of CE in dogs”</li> </ul>	<p><b>Symposium 12:</b> Neotropical Echinococcosis: emerging zoonosis</p> <p><u>CHAIR</u> Katherina Vizcaychipi, MSc (Argentina)</p> <ul style="list-style-type: none"> <li>- Katherina Vizcaychipi MSc (Argentina) “Epidemiological situation and importance of the Neotropical Echinococcosis in America and the world”</li> <li>- Katherina Vizcaychipi, MSc (Argentina) “Treatment of polycystic hydatidosis”</li> <li>- Natalia Casas (Argentina) “Neotropical and Cystic Echinococcosis, a multidisciplinary view from the public health perspective”</li> </ul> <p><u>SCIENTIFIC ORAL PRESENTATIONS</u></p> <p>33. Jhon Bosmediano (Peru)</p>	<p>Poster session 3 (Posters from 63 – 91)</p>
14:30 – 16:00	<ul style="list-style-type: none"> <li>- Graciela Santillán, Dr (Argentina) “Cystic Echinococcosis: environmental diagnosis”</li> </ul> <p><u>SCIENTIFIC ORAL PRESENTATIONS</u></p> <p>32. Luis M. Jara (Peru) “Development and validation of a Copro-ELISA sandwich for detection of Echinococcus granulosus-soluble membrane antigens in dogs”</p>		

		<i>“Neotropical Polychistic Echinococcosis in Cuniculus paca and Dasypus novemcinctus in the upper Itaya river basin, Peru”</i>	
Oct. 31  16:00 – 18:30	<p><b>Scientific Session 3 (oral presentations)</b></p> <p><u>CHAIR:</u> Leonardo Uchiumi, MD (Argentina)</p> <p>34. Malika Kachani (USA) <i>“Control of Hydatid Disease in the Context of the Neglected Zoonotic Disease System: Applying One Health in the Oloisukut Conservancy, Narok, Kenya”</i></p> <p>35. Veronica Poggio (Argentina) <i>“Eg95 vaccine monitoring of intermediate hosts. Analysis and inclusion of the socio-cultural context in vaccination programs”</i></p> <p>36. Francesca Tamarozzi (Italy) <i>“Epidemiological factors associated with human cystic echinococcosis investigated through a semi-structured questionnaire during the ultrasound surveys of the heracles project”</i></p> <p>37. Edmundo Larrieu (Argentina)</p>	<p><b>Scientific Session 4 (oral presentations)</b></p> <p><u>CHAIR:</u> Nazmiye Altintas, PhD (Turkey)</p> <p>44. Iris Fischer (Germany) <i>“Distribution of alveolar echinococcosis according to environmental and geographical factors in Germany, 1992-2018”</i></p> <p>45. Haihua Bao (China) <i>“Quantitative Study of Fat Deposition and Iron Deposition in Patients with HAE by HISTO and q-DIXON Techniques on MRI”</i></p> <p>46. Julian Schmidberger (Germany) <i>“Health-related quality of life in patients with alveolar echinococcosis: surgical versus conservative drug therapy – a quality of life comparison”</i></p> <p>47. Meng Qing Yang (China) <i>“Clinical Research of Shear Wave Elastography (SWE) in Differential</i></p>	<p>Poster session 3 (Posters from 63 – 91)</p>

	<p><i>“Identification of potential 'hot spots' of cystic echinococcosis transmission in the province of Río Negro, Argentina”</i></p> <p>38. Gulnara Minbaeva (Kyrgyzstan) <i>“Human Echinococcosis, Kyrgyzstan, 2001–2018”</i></p> <p>39. Katherina Vizchaychipi (Argentina) <i>“Cystic Echinococcosis in cattle of Misiones, Argentina”</i></p> <p>40. Maria Puchulu (Argentina) <i>“Social Representations on Echinococcosis cystic of families of High Mountain, Tucumán, Argentina”</i></p> <p>41. David Jenkins (Australia) <i>“Economic impact of hydatid disease on the Australian beef industry”</i></p> <p>42. Karima Achour (Algeria) <i>“Lung hydatid cyst of children”</i></p> <p>43. Karima Achour (Algeria) <i>“The specificity of the surgical management of the lung hydatid cyst”</i></p>	<p><i>Diagnosis of Hepatic Alveolar Echinococcosis (HAE) and Hepatocellular Carcinoma (HCC)”</i></p> <p>48. Bastian Marquis (Switzerland) <i>“Echinococcus multilocularis infection in solid-organ transplant patient recipients”</i></p> <p>49. Frantisek Stejskal (Czech Republic) <i>“Human Alveolar Echinococcosis in the Czech Republic: Clinical Outcome and Laboratory Parameters”</i></p> <p>50. Teivi Laurimäe (Switzerland) <i>“Long-term cryopreservation of Echinococcus multilocularis”</i></p> <p>51. Sven Baumann (Germany) <i>“Worldwide literature on epidemiology of human alveolar echinococcosis: a systematic review of research published in the twenty-first century”</i></p>	
18:30 – 19:00	<p><b>Closing Ceremony</b> <b>Organizing Committee of the 28<sup>th</sup> World Congress of Echinococcosis</b></p>		

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## CHAIRS AND SPEAKERS

1. Abela-Ridder, Bernadette, PhD (Department of the Control of Neglected Tropical Diseases, World Health Organization, Geneva, Switzerland)
2. Acosta, Gerardo, PhD (Instituto de Medicina Preventiva Veterinaria y Programa de Investigación Aplicada en Fauna Silvestre, Facultad de Ciencias Veterinarias, Universidad Austral de Chile, Chile)
3. Akhan, Okan, MD, Prof. (Faculty of Medicine, Hacettepe University, Ankara, Turkey)
4. Altintas, Nazmiye, PhD (Professor of Ege University, Medical Faculty, Department of Parasitology, Bornova-Izmir, Turkey)
5. Alvarez, Cristian, MSc (Institute of Parasitology, Vetsuisse Faculty, University of Zürich, Switzerland)
6. Brunetti, Enrico, MD (Department of Clinical Surgical Diagnostic and Paediatric Sciences, University of Pavia, Pavia, Italy)
7. Budke, Christine, PhD (College of Veterinary Medicine & Biomedical Sciences, Texas A&M University, USA)
8. Bustamante, José, DVM (Ministerio de Salud, Perú)
9. Casas, Natalia, Mg (Coordinación de Zoonosis, Ministerio de Salud y Desarrollo Social de la Nación de Argentina)
10. Casulli, Adriano, PhD (Department of Infectious Diseases, Istituto Superiore di Sanita', Rome, Italy)
11. Deplazes, Peter, Dr. (Institute of Parasitology, University of Zurich, Switzerland)
12. Elissondo, Celina, Dr (Laboratorio de Zoonosis Parasitarias, Instituto de Investigaciones en Producción Sanidad y Ambiente, Facultad de Ciencias Exactas y Naturales, Universidad Nacional de Mar del Plata, CONICET, Argentina)
13. Ferreira, Ciro, MD (Presidente de la Comisión Nacional de Zoonosis, Uruguay)
14. Flores, Ubaldo, DVM, Mg (Servicio Nacional de Sanidad Agraria, Ministerio de Agricultura y Riego, Peru)
15. Gavidia, César, DVM, MPH, PhD (Facultad de Medicina Veterinaria, Universidad Nacional Mayor de San Marcos, Peru)
16. Graeter, Tilmann, PD Dr. med. (Department of Diagnostic and Interventional Radiology, University Hospital Ulm, Ulm, Germany)
17. Grüner, Beate PD Dr.med. (Leitung Sektion Klinische Infektiologie Universitätsklinikum Ulm, Germany)
18. Guarnera, Eduardo, MD (Secretario General de la Asociación Internacional de Hidatidología/World Association of Echinococcosis, Argentina)
19. Heath, David, PhD (AgResearch New Zealand Limited, HopKirk Research Institute, Grasslands Research Centre, Palmerston North, New Zealand)
20. Jenkins, David, PhD (School of Animal and Veterinary Sciences, Charles Sturt University, Wagga, Australia)
21. Junghanss, Thomas, MD (Heidelberg University Hospital, Germany)
22. Kachani, Malika, DVM, PhD, NAPF (Professor of Parasitology & Global Health, College of Veterinary Medicine, Western University of Health Sciences, Pomona, California, USA)
23. Lightowlers, Marshall, PhD (University of Melbourne, Australia)
24. Larrieu, Edmundo, PhD (Universidad Nacional de Rio Negro, Argentina)

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25. Millon, Laurence, PhD, Prof. (Head of Parasitology-Mycology, University Hospital, Besançon, France)
  26. Montalvo, Raúl, MD, Mg, Dr (Jefe del Departamento de Enfermedades Infecciosas Tropicales del Hospital Daniel Alcides Carrión-Huancayo, Peru)
  27. Moro, Pedro, MD, MPH (Centers for Disease Control and Prevention, Atlanta, USA)
  28. Müllhaupt, Beat, Dr. Prof. (UniversitätsSpital Zürich Klinik für Gastroenterologie und Hepatologie, University of Zurich, Switzerland)
  29. Náquira, César, Dr (Universidad Nacional Mayor de San Marcos, Peru)
  30. Ran, Zhiguang, PhD, Professor (General Manager Assistant / Director of Quality, Research & Development Chongqing Auleon Biologicals Co., Ltd, China)
  31. Romig, Thomas, PhD (Universität Hohenheim, Parasitology Unit, Stuttgart, Germany)
  32. Santillán, Graciela, Dr (Departamento de Parasitología Instituto Nacional de Enfermedades Infecciosas -ANLIS Carlos G Malbran Argentina)
  33. Santivañez, Saul, MD, MPH, PhD (Universidad Peruana Cayetano Heredia, Peru)
  34. Sayes, Julio, MSc (Coordinador Técnico de la Comisión Nacional de Zoonosis, Uruguay)
  35. Siles, Mar, PhD (Instituto de Recursos Naturales y Agrobiología de Salamanca, Spain)
  36. Stojkovic, Marija, MD (Clinical Tropical Medicine Unit, Heidelberg University Hospital Germany)
  37. Tamarozzi, Francesca, PhD (Department of Infectious Diseases, Istituto Superiore di Sanita', Rome, Italy)
  38. Torgerson, Paul, PhD (Professor of Veterinary Epidemiology, Vetsuisse Faculty, Zurich, Switzerland)
  39. Uchiumi, Leonardo, MD (Ministry of Health of Río Negro Province, Argentina)
  40. Verastegui, Manuela, Mg, PhD (Universidad Peruana Cayetano Heredia, Peru)
  41. Vizcaychipi, Katherina, MSc (INMeT e INEI - ANLIS "Carlos G. Malbran", Argentina)
  42. Vuitton, Dominique A., MD, PhD (Professor Emeritus of Clinical Immunology, WHO Collaborating Centre on Prevention and Treatment of Human Echinococcosis, University Bourgogne Franche-Comté, France)

ROUND TABLE 1

Cyst Echinococcosis in Peru: Current Situation

Cystic echinococcosis in Peru: Current projects in CGH  
Saúl Santivañez  
Universidad Peruana Cayetano Heredia, Lima, Peru

Abstract is not available.

Contributing to control Cystic Echinococcosis  
César Gavidia  
[cgavidiac@unmsm.edu.pe](mailto:cgavidiac@unmsm.edu.pe)  
Facultad de Medicina Veteirnaria, Universidad Nacional Mayor de San Marcos  
Lima, Peru

Cystic Echinococcosis (CE) is still highly prevalent in Peru, particularly in the Central and South Highlands. Among the South American countries, Peru has the highest number of human CE cases. Several risk factors are present in those endemic areas, such as poverty, lack of sanitation, informal slaughterhouses, culling sheep in the backyard among others. Our research group are currently working on three different topics. First, sheep chemotherapy using oxfendazole (OXF), not as an animal model for human treatment but also as an additional strategy to reduce the protoscolex viability in naturally infected sheep. The efficacy of OXF has been demonstrated in a serie of field clinical trials in sheep; from the initial treatment scheme of 30mg/Kg weekly x 11 weeks this has been reduced to 60mg/Kg weekly x 4 weeks. The dose has been increased while the time has reduced, making the treatment more practical. Although OXF efficacy has not reached 100%, the reduction of protoscolex viability has been significant. The second research line is the immunization of dogs against *E. granulosus*. Our preliminary results demonstrated that excretory/secretory antigens as well as membrane proteins might trigger the immune system in vaccinated dogs, reducing the number of worms as compared to the control group. However, this research area still needs further studies to significantly conclude the utility of the vaccine in dogs. Finally, we have stardized an *in house* copro ELISA test, which was published a year ago. This test has a sensitivity of 96% and a specificity of 98%, with no cross reactions with other parasites such as *T. hydatigena*, *Toxocara canis*, *D. caninum*. This test is being used in the Pilot Control Program in Peru to estimate the prevalence and incidence in dogs in that endemic area. Future studies to evaluate efficiency of this copro ELISA are needed.

The Control and Prevention of Hydatidosis/Cystic Echinococcosis Pilot Program in Peru  
Ubaldo Flores<sup>1,2</sup>, Juan Calcina<sup>1</sup>, Miguel Quevedo<sup>1</sup>

<sup>1</sup> National Service of Agrarian Sanity (SENASA), Ministry of Agriculture, Lima, Peru. <sup>2</sup>  
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In 1974 a Pilot control program to Cystic Echinococcosis (CE) elimination was developed in Junin – Peru by Ministry of Health and Ministry of Agriculture in collaboration to others

Public and private institutions, the social conflicts forced to stopping the pilot program in the early 80's. After the failing of that pilot control program, there was not any other attempt to control CE in Peru. In 2013 the Health Ministry and Agriculture Ministry supported that the government of Peru declared the National Interest for Surveillance, Prevention and Control of Cyst Echinococcosis/Hidatidosis, the declaratory emitted by the government of Peru appointed a multi sectorial committee was assigned to release guidelines protocols for Hydatidosis prevention and control program. By this guidelines The National Service of Agrarian Sanity (SENASA by his acronyms of Spanish) started the The Control and Prevention of Hidatidosis/Cystic Echinococcosis Pilot Program in 2015. The aim of this pilot program is the establishment the effectiveness and the most economically viable strategies to be recommended in a future national program. Five regions of Peru were selected for the Control Program (Cusco, Junin, Huancavelica, Pasco and Puno), a total of seventeen rural communities from those regions were chosen as the pilot areas for the control program. In order to measure the outcome of the strategies, the program is monitoring canine echinococcosis by means of copro ELISA test, cystic echinococcosis in sheep using serological assays such as Western blot (P L Moro, Verastegui, et al., 1997). The pilot control program is evaluating four different strategies which were implemented either alone or in combination among the selected communities: Dog Antiparasitic treatment (Praziquantel 5mg/kg), Oxfendazole sheep treatment (60 mg/kg) sheep vaccination by EG95 vaccine and educational program in all rural communities. The total duration of the Pilot program is five years and it finalize in 2019.

Key words: Hidatidosis, Cystic Echinococcosis, Pilot Program, oxfendazole, praziquantel, EG95.

**CYSTIC EQUINOCOCCOSIS IN PERU  
M.V. JOSE LUIS BUSTAMANTE NAVARRO**

National Coordinator of Zoonoses - Directorate of Prevention and Control of Metaxenic Diseases and Zoonoses - General Directorate of Strategic Public Health Interventions.  
Ministry of Health of Peru

Hydatidosis is a parasitic zoonosis, a disease produced by the larval state of the Echinococcus cestode of the genus Granulosus, of high prevalence in our country, which is mainly registered in the central highlands, this mainly due to the habit of living with dogs and sheep as well as the lack of education and awareness of the population. But also unlike other countries, our sheep farming is mainly concentrated at the level of small producers, where approximately 60% sheep are Creole, 25% crossed and 15% breed, with very low production and productivity averages. The importance of this zoonosis in public health is related not only to the high rate of human morbidity and mortality, but also to losses due to work performance, hospitalization expenses, interventions and disabilities. The economic loss due to human hydatidosis according to studies has been estimated at 850,000 dollars and in the livestock economy approximately 500,000 dollars per year, which would reflect losses greater than 1,000,000 dollars per year. The high economic losses at the livestock level, due to the system of our livestock are not perceived by the breeders, who commercialize mostly in the field or benefit for self-consumption and home sales. This makes awareness and application of knowledge difficult to prevent hydatidosis at the population level, especially in rural areas. Many pilot programs have been developed in the country, finding in some of them the feasibility of controlling this zoonosis, especially where there is organized livestock. In recent years in the country the diagnosis of human hydatidosis has been improved, both by the mplementation of imaging and serological diagnostic

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techniques, which are being embodied in the normative document in the approval process. Peru has a decentralized health care system, managed by five entities: the Ministry of Health (MINSa), which offers health services for 60% of the population; EsSalud, which covers 30% of the population; and the Armed Forces (FFAA), the National Police (PNP), and the private sector, which provide health services to the remaining 10%. According to the existing reports between 2002 and 2018, the Ministry of Health reported 54,695 cases of which 21992 cases (41%) in the capital of Peru and EsSalud between 2013 - 2018 reported 10422 cases, of which 2671 (26% ) were reported in Lima, due to the level of complexity and resolving capacity of health facilities. In the Ministry of Health in 2018, 4794 cases were reported. The average prevalence rate in Peru is 12 cases x 100, 000 inhabitants, with regions such as Pasco, Junín, Huancavelica, Puno and Lima, where they report between 14 - 34 cases per 100,000 inhabitants, but in almost the entire country there is Some kind of regional report.

## Symposium 1

### Imaging methods in the diagnosis and follow up of human Cystic Echinococcosis

US, CT and MRI in cystic echinococcosis  
Thomas Junghanss, MD

Abstract is not available.

US in follow-up (watch and wait) overview of evidences gathered so far  
Marija Stojkovic, MD

Abstract is not available.

Update on the WHO-IWGE work on imaging  
Francesca Tamarozzi, PhD

Abstract is not available.

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## Symposium 2

# Molecular biology of the species of *Echinococcus*

Species and genotypes: what is new?

Thomas ROMIG<sup>1</sup>, Marion WASSERMANN<sup>1</sup>

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According to most recent reviews on *Echinococcus* taxonomy, the genus is composed of nine different species, five of them belonging to the *E. granulosus* sensu lato cluster. However, in some cases the intraspecific variance is poorly known, and two of the species with the highest relevance for human health show a complex genetic structure. For *E. granulosus* sensu stricto, two major genotypes are proposed (G1 and G3), who are closely related and globally distributed; differences concerning pathogenicity and host predilection have not been convincingly demonstrated. An additional, very divergent genotype, G-Omo, is present in eastern Africa and may – based on genetic distance from G1/3 – in future have to be separated from *E. granulosus* s.s. The most diverse species is *E. canadensis*, whose three major genotypes (G6/7, G8, G10) are clearly separated concerning genetic distance, geography and host range. Subdivisions into two or three species have been proposed, but a consensus on names has not yet been reached due to gaps of knowledge. Here we give a concise overview on the current taxonomic concepts with accepted and debated issues and highlight the relevance of *Echinococcus* taxonomy for epidemiology and public health.

Keywords: *Echinococcus*, taxonomy, species, genotypes

### **SCIENTIFIC ORAL PRESENTATIONS**

Distribution map of *Echinococcus granulosus* genotypes in Turkey

Mesut AKIL<sup>1</sup>, Ozge SARICA YILMAZ<sup>2</sup>, Eylem AKDUR OZTURK<sup>1</sup>, Nuray ALTINTAS<sup>2</sup>, Aysegul UNVER<sup>1</sup>, Nazmiye ALTINTAS<sup>1</sup>

<sup>1</sup>Ege University, Faculty of Medicine, Department of Parasitology, Izmir, TURKEY,

<sup>2</sup>Manisa Celal Bayar University, Faculty of Medicine, Department of Medical Biology, Manisa, TURKEY

Introduction: Echinococcosis is a parasitic infectious disease caused by *Echinococcus granulosus* which is cryptic species complex. This complex is intermixed of *E. granulosus sensu stricto* (G1-G3), *E. equinus* (G4), *E. orteppi* (G5), and *E. canadensis* (G6-G10). This parasite can infect wide range of domestic and wild mammals as well as humans in endemic countries like Turkey. The aim of this study is to provide summary information about the distribution of *E. granulosus* genotypes which is detected with molecular methods in human and animal isolates in Turkey until now. Material and methods: The available articles were identified by searching electronic international databases (PubMed, Google Scholar and Science direct) and national databases (Turk Medline, ULAKBIM) that detected genotypes in Turkey or Turkish origin isolates. All data were entered into table and prepared map of detected genotypes. Results: A total of 1884 studies were identified by searching. Of these, a total of 42 were identified as eligible papers which include 1692 human and animal samples. Genotype G1 was the most prevalent with rate 61.2% (95% CI: 50.2%-72.1%). After that rates were detected as; genotype G1-G3 was 29.3% (95% CI: 18.8%-39.7%) , genotype G6/G7 was 0.3% (95% CI: 0%-

1%), genotype G7 was 0.5% (95% CI: 0%-1.2%), genotype G4 was 4% (95% CI: 0%-9%), genotype G3 was 4.22% (95% CI: 0.1%-8.3%), genotype G6 was 0.9% (95% CI: 0%-0.3%). Conclusion: With this study, genotype G1 is most predominant genotype in Turkey and it infects various mammals (cattle, sheep, goats, dogs, buffalo etc.) and human. In addition, it was shown the regions of *E.granulosus* genotyping in Turkey. Thus, it will be a guide for where genotyping studies have not been performed in the other regions of the country.

European Alveolar echinococcosis genotyping: an EmsB microsatellite analysis  
Jenny Knapp <sup>1,2</sup>, Bruno Gottstein, Stéphane Bretagne, Jean-Mathieu Bart, Gérald Umhang, Carine Richou, Solange Bresson-Hadni, Laurence Millon.

<sup>1</sup> UMR CNRS 6249 Chrono-environnement, Université Franche-Comte, <sup>2</sup> Parasitology-Mycology, University Hospital of Besançon, 25030 Besançon, France. [jknapp@chu-besancon.fr](mailto:jknapp@chu-besancon.fr); [lmillon@chu-besancon.fr](mailto:lmillon@chu-besancon.fr)

Introduction: Associating the genetic diversity of *Echinococcus multilocularis* to well-documented contamination events is, for specialists, similar to questing of the grail. Indeed, tracing back the history of contamination i.e. exposure to infection in patients suffering from alveolar echinococcosis (AE) is often a difficult task because of the longterm development of the parasite in the human liver and the belated nature of the diagnosis. Moreover, the occurrence of different clinical forms in humans (extra-hepatic locations, ability to develop metastases, etc.), raises the issue of pathogenicity levels in *E. multilocularis*, e.g. in relation to the exposition of humans to different strains or genotypes putatively circulating in the environment. Material-Methods: Among presently available molecular tools, the EmsB microsatellite was largely studied during this last decade, and a collection of 1,211 European reference samples has been accumulated so far. A panel of 63 surgical samples was collected between 1981 and 2018 from AE-patients (31 women and 31 men, from 16 to 80 years old, median age 60 years old) originating from five European countries (France, Switzerland, Germany, Belgium and Luxembourg). Results: The present study allowed us to distinguish eight EmsB profiles, four of them were shared among patients located in the same area of France or Switzerland, one profile was shared on both sides of the French-Swiss border, while patients from non-highly-endemic regions were clustered together in one profile. Extra-hepatic lesions shared, for some of them, the same profile, raising the question if these patients got infected with peculiar *E. multilocularis* profiles. EmsB profiles also seemed to circulate on a large time period because patients with ancient or recent surgical resection partially shared similar profiles. Conclusion: This present study allows us to put into perspective the contamination pathways in humans, based on proximity contamination in some cases and contamination with dominant profiles in Europe, especially for extra-hepatic lesions.

Probability of detection of *Echinococcus multilocularis* DNA in fox fecal samples  
Laure Bournez, Alice Schollhammer, Franck Boué, Gérald Umhang.  
[gerald.umhang@anses.fr](mailto:gerald.umhang@anses.fr)

ANSES, Nancy Laboratory for Rabies and Wildlife, Malzéville, France

The detection of *Echinococcus multilocularis* DNA in fecal sample of definitive hosts is nowadays routinely used. Nevertheless, little attention is paid to the representativity of the portion taken in relation to the whole fecal sample. In order to evaluate the homogeneity of *E. multilocularis* DNA presence in fecal sample, 75 stools of 8 foxes

infested experimentally taken at different period of infestation and 45 stools of naturally infested foxes were tested by real-time PCR five times by portion of 500mg each. Experimentally, 76% (13/17) and 60% (3/5) stools taken respectively during the beginning of the prepatent period and during the patent period was positive at least 4 times over 5. This proportion was much lower for the prepatent period (24%). For the feces from naturally infected foxes, 42% were systematically positive for the five portions taken when 20, 18 and 13% of the feces were positive 4, 3 and 2 times, respectively. The variation of intra-stool Ct-value increased with the mean Ct-value of the stool, with a median difference of 5 Ct for stools having a mean Ct-value of 30-35 and of 35-40. By using data on naturally infected foxes, we estimated the probability for detecting DNA of *E. multilocularis* from an infected fecal sample increased from 75% to 95% when one to five portions of fecal sample is tested. For experimentally infected foxes, the probability of DNA detection when one portion of stool is tested was 92% during the beginning of the prepatent period, 51% during the prepatent period and 75% during the patent period. The heterogeneity of the *E. multilocularis* DNA in fecal samples highlighted in this study should be considered for individual diagnostic improvement by testing several portions but also for the surveillance context by adjusting the sampling size and the confidence interval associated to the prevalence obtained.

## Symposium 3

### Immunological methods of the diagnosis and follow-up of human Cystic Echinococcosis

Challenges to improve Serologic Diagnosis of Cystic Echinococcosis  
Manuela Verastegui, PhD  
Universidad Peruana Cayetano Heredia, Lima, Peru

Abstract is not available.

#### **SCIENTIFIC ORAL PRESENTATIONS**

Diagnostic performances of commercial elisa, iha, and wb for the diagnosis of echinococcal vs non-echinococcal lesions: a retrospective analysis of data of a single reference centre

Ambra VOLA, Tommaso MANCIULLI, Raffaella LISSANDRIN, Mara MARICONTI, Mar SILES-LUCAS, Enrico BRUNETTI, Francesca TAMAROZZI

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**INTRODUCTION:** The diagnosis of cystic echinococcosis (CE) is based on the observation on imaging of the pathognomonic features of the parasitic cysts. Serology has a role in the differential diagnosis of echinococcal vs non-echinococcal lesions when imaging is inconclusive but there is no consensus on the diagnostic algorithm to apply. We

performed a retrospective analysis of data on three serological tests applied to patients with untreated hepatic CE cysts and non-CE lesions entering in differential diagnosis, visited in the outpatient service of San Matteo Hospital Foundation, Pavia, Italy. MATERIALS and METHODS: Data were retrieved for eligible patients diagnosed between 2005-2017 who were tested with the same three commercial serology assays: ELISA RIDASCREEN Echinococcus IgG (R-biopharm), IHA Cellognost Echinococcosis (Siemens), and Western Blot Echinococcus WB IgG (LDBIO Diagnostics). Diagnostic performance characteristics were calculated for each test and their combinations, on the whole cohort and on separated groups ("active CE vs liquid non-CE" and "inactive CE vs solid non-CE" lesions). RESULTS: 104 patients with liver CE (52 with active and 52 with inactive cysts) and 257 patients with non-CE hepatic lesions (245 liquid and 12 solid) were included. WB showed the best diagnostic performances as a single test or as a confirmatory test of ELISA+IHA discordant/negative results. The diagnostic performances of the combination of tests will be presented. DISCUSSION/CONCLUSIONS: The diagnosis of CE cases can be challenging. Ultrasonography is operator-dependent; therefore, its diagnostic accuracy may vary considerably with the operator. Furthermore, the prevalence of CE and the rate of CE cysts vs non-CE lesions diagnosed with ultrasound in a population influence the pre-test and post-test probabilities of a lesion being CE. Our work, although retrospective, provides data in support of the development of a consensus algorithm for the serology-integrated diagnosis of CE, to be tested prospectively in different settings.

Serum Ferritin combined with MRI in identifying IgG negative Application study of hepatic cystic echinococcosis and hepatic cysts

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Objective: To investigate the diagnostic value of serum ferritin in IgG-negative CE1 hepatic cystic echinococcosis and hepatic cysts, and to explore the ability of serum ferritin to distinguish the two diseases. Materials and Methods: A retrospective analysis was performed on the demographic, clinical and imaging data of 80 patients with IgG-negative CE1 hepatic cystic echinococcosis and 60 patients with simple hepatic cyst diagnosed and treated by Hepatobiliary and Pancreatic Surgery, Affiliated Hospital of Qinghai University from 2016 to 2018. The relationship between serum ferritin and other indicators was summarized. The value of MRI combined with serum ferritin in the differential diagnosis of hepatic cystic echinococcosis and simple hepatic cyst was summarized. Results: The average age of 80 patients with IgG-negative CE1 hepatic cystic echinococcosis was  $37.04 \pm 11.07$ , and the average age of 60 patients with simple hepatic cyst was  $51.38 \pm 11.10$ . The average diameter of hepatic cystic echinococcosis and simple hepatic cyst was  $5.88 \text{ cm} \pm 1.88 \text{ cm}$ ,  $2.89 \text{ cm} \pm 1.28 \text{ cm}$ ; SF index  $P < 0.05$ , SF index of liver cystic echinococcosis was  $450.02 \pm 350.85 \text{ ng/ml}$ , simple liver cyst SF index was  $204.38 \pm 159.24 \text{ ng/ml}$ ; Differential analysis of hepatic cystic echinococcosis and simple hepatic cysts: Fe-, AFP, CA125, CA199, ALT, ALP, TBA, There were statistical differences in TG and HDL indexes,  $P < 0.05$ ; elevated SF index in patients with hepatic cystic echinococcosis and AFP, ALP, GGT, jaundice indicators (DBIL, IBIL). At the same time, the long-path of the lesion was the most correlated with the increase of SF; However, SF elevation in patients with simple hepatic cyst was only correlated with CEA and lesion diameter. Conclusion: There is a significant difference in SF content between CE1 type hepatic cystic echinococcosis and

simple hepatic cyst. Combined with SF index, it will improve the discrimination ability of CE1 type IgG negative hepatic echinococcosis and simple hepatic cyst, and provide reference for clinical treatment value.

Key words: MRI; Serum Ferritin; Hepatic Cystic Echinococcosis; Hepatic Cystic; Differential Diagnosis;

Fund Project: Popularization and Application of Remote Image Diagnosis Technology in Rural Pastoral Areas of Qinghai Province (2017-SF-158).

Value of soluble programmed death-1 (sPD-1) and sPD-ligand-1 (sPDL-1) as immunological biomarkers for the follow-up of cystic echinococcosis in Tunisian pediatric patients

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**Introduction.** Cystic echinococcosis (CE), caused by *Echinococcus granulosus* continues to be a public health issue in developing countries including Tunisia. Available serological immunoassays show unsatisfactory performance for the follow-up of CE so the development of reliable tests is highly demanded. The detection of immunological markers such as immune-checkpoints may be an interesting alternative to monitor primary cure and detect relapses. Here, we investigated the potential role of soluble programmed death-1(sPD-1)/sPD-ligand-1 (sPDL-1) pathway in controlling the CE infection by measuring the sera levels of sPD-1 and sPDL-1. **Methods.** This study was carried out on 72 sera collected from 24 Tunisian pediatric patients who were operated for exclusively pulmonary CE and followed-up during 1 post-operative year. For each patient, sera samples were obtained at three points of time. Patients were classified into two groups based on post-surgical outcomes “the cured” CE (CCE) patients and “non-cured” CE patients (NCCE). Serum concentration of sPD-1 and sPDL-1 were measured using ELISA commercial kits (Invitrogen, Thermo Fischer Scientific, Austria). We used the Wilcoxon test to compare the levels of sPD-1 and sPDL-1 between the three points of time in the CCE and NCCE groups. **Results.** Our results show a significant decrease of sPDL-1 expression levels one year after surgical treatment compared to pre-operative rates in CCE group ( $p < 0.005$ ), however in NCCE patients no significant changes were observed during all the follow-up period. The comparison of sPD-1 sera rates measured 1 year post-surgery to expression levels in 1 month in CCE showed a marginal trend toward significance ( $p = 0.060$ ), while NCCE group exhibited persistent high sPD1 levels during all the 12-months of the follow-up period. **Conclusion.** This study showed that monitoring the sPD-1/ sPDL-1 sera levels in patients with pulmonary CE may be a potential attractive strategy to evaluate the effectiveness of surgical treatment and detect eventual recurrences.

Key words: sPD-1/sPDL-1, Follow-up, Cystic echinococcosis

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## Symposium 4

### Diagnosis and treatment / prevention of cystic echinococcosis (intermediate hosts, animal models)

Can pharmacotechnical strategies be useful to optimize the anti-echinococcal efficacy of albendazole?

María Celina ELISSONDO

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In the last 30 years, an increase in the use of anthelmintic drugs for the medical treatment of human cystic echinococcosis was observed. Moreover, chemotherapy strategies can be added as an additional control measure in animals (intermediate hosts). Albendazole has limited efficacy due to its poor and erratic bioavailability, attributed to its low dissolution rate. On the other hand, the poor water solubility of albendazole offers only few formulation possibilities, limiting the administration routes. Modern pharmaceutical technology may help to greatly improve albendazole systemic exposure improving its efficacy against the parasite. In this presentation, some of the new technologies applied to the experimental treatment of echinococcosis will be discussed. During the last 20 years, the Parasitic Zoonoses Research Group (Faculty of Natural and Exact Sciences, Mar del Plata National University, CONICET) has been working in the experimental chemotherapy of cystic echinococcosis. Since 2011, in a joint effort with the Laboratory of Pharmacotechnics (Faculty of Chemistry, National University of Cordoba, CONICET), different albendazole drug delivery systems are being studied with the aim to improve the treatment against the murine model of echinococcosis. After evaluating the clinical and chemoprophylactic efficacy of two solid dispersions, nanocrystals and lipid nanoparticles, promising results have been obtained.

EG95 Vaccine Field Trials in China, New Zealand Handover Document, Production in China Factory, Safety and Efficacy to Registration.

HEATH David, AgResearch New Zealand ([heathdd@gmail.com](mailto:heathdd@gmail.com)) and LIGHTOWLERS Marshall, University of Melbourne, Australia, FU Gang, RAN Zhiguang, ZHAO Yangyang, WANG Chengzhuo, CHEN Qianlin, LV Hang, GU Jing, LI Yangchun, Zheng Cao, Jane Jian, Mei Song. Chongqing Auleon Biologicals Co. Ltd, Chongqing 402600, AND Chi Pusheng, Shi Baoxing, Zeng Zhuangzhi, Huang Yan, Wang Jincheng. China and Engineering Technical Center of Biologics, Chinese Academy of Agricultural Sciences, Peking, 100081

Introduction: Describes the Field Trial of New Zealand Registered EG95 vaccine sent to China during the years 1996-1998 and then the New Zealand Hydatids Vaccine Handover Document given to the Chinese Ministry of Agriculture, Department of Livestock Husbandry. Material and Methods: From the beginning of 1996 to the end of 1998, a field trial of Echinococcosis recombinant subunit vaccine (sheep & goats) was set up in Xinjiang Province for 4 years. More than 3000 sheep were vaccinated, some of which were challenged with *E. granulosus* eggs. Protection trial and Longevity Test of Immunity were carried out. 354 vaccinated sheep were necropsied. Cyst reduction rate reached to 81~99%, the immunity could last more than 1 year. From March, 1996 to June, 1997 we

carried out the first phase of the trial, together with the veterinary station, clinics, counties and farms in Aksu Prefecture and Bayingol Mongolian Autonomous Prefecture of Xinjiang Uygur Autonomous Province of China. The following 9 procedures were needed to be done: selection of trial locations, preparation of *E.granulosus* eggs by artificial infection of domestic dogs, grouping of test animals, blood sampling and preparation of sera, vaccination, artificially challenge, determination of antibody level of sera by ELISA assay, necropsy of test sheep, examination of cysts in liver and lung and evaluation of results.

Results: We fulfilled the mission and achieved the expected objective. The cyst reduction rate of the two trials both achieved more than 80%. Discussion/Conclusion: The Handover document from New Zealand was passed to China in 2000. By 2004 a factory was built in Beijing by CAAS and large amounts of quality vaccine was produced. This vaccine was then tested in Qinghai and Xinjiang. Good results meant that the vaccine was registered in China in 2007. A new factory to GMP standards was set up in Chongqing Auleon Biological Co. Ltd in 2008. In 2009 obtained GMP certificate. In 2011-2015 Auleon Biologicals conducted pilot field trials of the vaccine in Xinjiang. 4.7 million doses were tested. Two kinds of ELISA kits were developed- EG95 to detect antibody after vaccination, and AgB to detect *E.granulosus* mature cysts.

Keywords: EG95 Vaccine, New Zealand, China.

## **SCIENTIFIC ORAL PRESENTATIONS**

A platform for identifying novel carbazole aminoalcohols against echinococcosis  
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**Objective:** Echinococcosis caused by the cestode *Echinococcus granulosus* and *E. multilocularis*, is a worldwide zoonosis. Our study is to establish an in vitro and in vivo platform for screening small molecules against echinococcosis. **Material and Methods:** Carbazole aminoalcohols were tested against *E. granulosus* protoscoleces in vitro and metacestodes ex vivo. The in vivo chemotherapeutic effect of representative compounds was also assessed in experimentally infected mice, and corresponding effects were visualized by transmission electron microscopy. **Results:** The carbazole aminoalcohols exhibited potent protoscolicidal activity with LC50 values ranging from 18.23 to 34.27  $\mu$ M. Among them, compounds #2 and #24 at a concentration of 12 mg/L killed all ex vivo cultured metacestodes. In vivo studies showed that oral administration of compounds #2 and #24 at a dosage of 25 mg/kg/day for 30 days led to reductions of 68.35 and 67.90% in parasite weight compared with the untreated group (both groups:  $p < 0.001$ ). Compound #2 at a dosage of 25 mg/kg/day and compound #24 at 50 mg/kg/day induced significantly higher cyst mortality rates in comparison with that of the albendazole group (both groups:  $p < 0.01$ ). Analysis of parasite tissue collected from compound #2- or #24-treated mice by transmission electron microscopy revealed a drug-induced complete structural destruction. The structural integrity of the germinal layer was totally lost, and the majority of the microtriches were absent. **Conclusions:** Our study identifies that carbazole aminoalcohols are novel agents against echinococcosis. Compounds #2 and #24 represent promising drug candidates in anti-echinococcosis chemotherapy.

**KEY WORDS:** cystic echinococcosis; alveolar echinococcosis; in vitro and in vivo models; inhibitors; drug targets; carbazole aminoalcohols

## TOWARDS AN EXPERIMENTAL MODEL FOR STUDYING CYST FORMATION

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**Introduction:** Cyst formation in echinococcosis/hydatidosis is a poorly studied process, due to lack of models other than experimental infection in secondary hosts. Those infections are ethically questionable, with elevated requirements of breeding, animal care and usually take months of follow-up. Mouse inoculations with protoescolices don't resemble parasite behavior occurring in secondary hydatidosis. In the '60 and '70 decades few reports about morphological description of cyst formation in culture were published. A couple of reports in 1990s have documented more features, but there are no molecular studies on this process. New tools as genomics, proteomics and metabolomics would allow studies in this area, but it's necessary to have alternative experimental models which overcome mentioned difficulties. The aim of present work was to perform a screening study to determine suitable preparations as new alternative models to study secondary cyst formation in hydatidosis/echinococcosis. **Materials and Methods:** Healthy and echinococcus infected cattle livers were obtained from slaughterhouse. The protoescolices from cysts of same animal were pooled, DNA extracted, PCR amplified and sequenced, in order to determine species and genotype. Fresh protoescolices were microscopically observed for vitality determination. Several liver preparations were in vitro cultured with DMEM, antibiotic supplemented, with and without protoescolices. Free liver preparations of protoescolices were cultured as controls. Follow up with daily microscopic observation within at least a week was performed. **Results:** After 48-72 hours of incubation a clear halo surrounded some evaginated protoescolices, compatible with enzymatic degradation of liver preparations. Those halos were not observed in control wells. **Discussion:** The features registered resulted compatible with extracellular matrix degradation by the protoescolices. Such degradation could be necessary for parasite to achieve cyst location site. Despite much work is still to be done, the results are promising and the molecular characterization on these models would allow studying aspects on this illness never pointed before.

**Keywords:** experimental model, secondary hydatidosis, cyst formation

## SERODIAGNOSTIC STUDY OF CYSTIC ECHINOCOCCOSIS IN SHEEP FROM AREAS OF AMAZONIAN DOMINION, PROVINCE OF MISIONES, ARGENTINA

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The province of Misiones in Argentina located in the amazon domain, presents the cystic echinococcosis (CE) with an identified dog-bovine cycle and neotropical echinococcosis with wild infection cycles. In the last six years an increase in ovine and caprine production, with 15.000 and 6.000 head respectively. It is observed that there is no availability of sheep and goat abattoirs in the area that provide information of CE in these animals. This work aimed is to perform epidemiological surveillance of CE in sheep through serodiagnosis and safe slaughter training for the staff of the evaluated farms. Descriptive design, blood samples were taken from 35 sheep from three different epidemiological units (EU), Fachinal and San José, Misiones and Caa Carai, Corrientes. The sampled animals were marked for follow-up. Race, category, sex, traceability and associated risk factors were recorded. Serum samples were analyzed by an IgG ELISA kit using a recombinant B8/2 subunit of antigen B of *Echinococcus granulosus* (99% Sensitivity-98% Specificity). Each producer gave his consent and commitment to report after the slaughter the presence of cysts in viscera, canine periodic deworming and destruction of their scat. The results showed 25,7% of sheep serologically positive. According to the category and traceability of the animal, in 2/3 EU positive lambs were observed (2,8% Misiones and 5,7% Caa-Carai). 17,1% (6/35) of the sheep with traceability from Misiones, Corrientes and Santa Fe were positive. Five trainings were carried out. These preliminary data constitute the first reported situation in sheep for Misiones. The use of serology allowed advancing and performing corresponding interventions from the animal health, to prevent future disease in people. According to the needs and difficulties of each region, the use of serodiagnosis as a tool for the surveillance of CE in intermediate host, would be a potential way forward.

Keywords: Cystic Echinococcosis - serodiagnosis - sheep - Misiones, Argentina

## Conference 1

### Socioeconomic impact of Echinococcosis in endemic countries

Overview of the Socioeconomic Impact of Echinococcosis in Endemic Countries  
Christine Budke, PhD (USA)

Abstract is not available,

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## ROUND TABLE 2

### Situation of Cystic Echinococcosis around the world

#### THE BURDEN OF CYSTIC AND ALVEOLAR ECHINOCOCCOSIS IN EUROPE

Adriano CASULLI

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**INTRODUCTION:** *Official burden:* Cystic (CE) and alveolar echinococcosis (AE) global prevalence is estimated at 1–3 million human cases. However, clinically diagnosed cases represent only a small proportion of the total number of real infected people. For instance, even if human AE and CE are notifiable in some European member states (MS), in practice, these parasitic diseases are largely underreported in Europe. In addition, cases of both AE caused by *E. multilocularis* and CE by *E. granulosus sensu lato (s.l.)* are reported jointly as “echinococcosis”, as the European Union (EU) case definition does not distinguish between these two parasitic diseases. *Hidden burden:* In this context, a cross-sectional study on abdominal prevalence of CE was undertaken in Eastern Europe (Romania and Bulgaria) and Turkey under the framework of HERACLES project to quantify asymptomatic carriers and allow a more precise estimate of CE burden. **MATERIALS and METHODS, and RESULTS:** *Official burden:* Around 700/800 human cases of “echinococcosis” are recorded every year in Europe. For instance, 827 confirmed human “echinococcosis” cases were officially reported in the EU with a notification rate of 0.19 cases per 100,000 population in 2017. Eleven MS reported data on 7,148 foxes examined for *E. multilocularis*, and nine MS reported positive findings with an overall prevalence of 16.9%. Data of 2017 from Finland, Ireland, Malta, the United Kingdom and Norway confirmed the free status of these countries for *E. multilocularis* in the context of Regulation (EU) 1152/2011. For *E. granulosus s.l.*, 21 MS reported data from around 111 million animals and nine MS reported positive samples with an overall official prevalence of 0.24% in 2017. *Hidden burden:* Within the framework of HERACLES project, 16 abdominal ultrasound surveys were conducted in 50 villages in association with resident partners and public health centres in Bulgaria, Romania and Turkey. 24,693 people were screened for CE during 2014 and 2015. The age and sex adjusted prevalence of abdominal CE was 0,41% (95%CI 0,29–0,58) in Bulgaria, 0,41% (95%CI 0,26–0,65) in Romania, and 0,59% (95%CI 0,19–1,85) in Turkey. Active cysts were found across all ages, including children, and in all investigated provinces. Based on the adjusted prevalence and the reference rural population size in 2015, it was estimated that 7,872 (95%CI 5,520–11,220) individuals may be presently infected with abdominal CE in rural Bulgaria, 37,229 (95%CI 23,405–59,166) in rural Romania, and 106,237 (95%CI 33,829–330,751) in rural Turkey. Of these, 42,9% in Bulgaria, 40,3% in Romania, and 32,8% in Turkey may harbour cysts in active stage. **CONCLUSION:** Collection of accurate epidemiological and clinical data will give a reliable picture of the burden of these diseases in Europe, providing a statistically supported case series for future evaluation of efficacy and effectiveness of interventions. With the aim of improving surveillance of CE and AE, we encourage international agencies to lobby the EC to champion new health policies for the notification of human and animal CE and AE. This work was supported by the EU FP7, HERACLES project (GA602051; <http://www.Heracles-fp7.eu/>) and European agencies ECDC (<https://ecdc.europa.eu/en/home>) and EFSA (<http://www.efsa.europa.eu/>).

**Keywords:** Burden of disease; Public health; cystic and alveolar echinococcosis.

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National Control Program of Cystic Echinococcosis in New Zealand. Cystic Echinococcosis in Australia. Cystic Echinococcosis in China  
David Heath, PhD (New Zealand)

Abstract is not available.

National Control Program of Cystic Echinococcosis in Uruguay: achievements and future challenges  
Julio Sayes, MSc (Uruguay)

Abstract is not available.

## Symposium 5

### Surgery and other intervention options for Cystic Echinococcosis treatment in humans

PAIR: An overview  
Enrico Brunetti, MD (Italy)

Abstract is not available.

Tratamiento quirúrgico selectivo de resección adventicial sin riesgo y desconexión quisto biliar en los Quistes Hidáticos Hepáticos  
Echeveste, José; Correa, Carlos; Abadie, Rafael; Ferreira, Ciro; Perdomo, Roberto (QEPD)  
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La adventicia de los quistes hidáticos del hígado evolucionados adquiere caracteres físicos de engrosamiento escleroso y calcificación que la hacen proclive a la infección y gravan los procedimientos operatorios que la abandonan total o parcialmente (quistectomía parcial). Ello da lugar a la propuesta de recurrir en todos los casos a la quistectomía total que elimina ese riesgo y crea otros. Es preciso discriminar el tipo de quistes que por aquellas características de su adventicia –y teniendo en cuenta también su tamaño y topografía- se prestan a la discusión entre su tratamiento "conservador" o "radical" o una combinación de los mismos. La infección posoperatoria de la adventicia residual tiene como causas principales la retención cavitaria en los procedimientos cerrados y la bilirragia cuando existe una comunicación quisto biliar. Si se evitan estas causales se puede tener una evolución con tolerancia de los restos de adventicia hidática en las quistectomías parciales. Lo que es perfectamente factible tan pronto se logre: 1) mantener la cavidad abierta hacia el peritoneo; 2) realizar la desconexión quisto-biliar para evitar bilirragia y estenosis; 3) evitar la colocación de tubos en cavidad cerrada; 4) administrar antibioticoterapia preventiva. Esto es el tratamiento quirúrgico selectivo de resección adventicial sin riesgo y desconexión quisto biliar en los quistes hidáticos hepáticos. En contrapartida se resaltan los riesgos inconvenientes de las quistectomías

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totales. Presentamos una serie de 320 pacientes sometidos a este tratamiento en estos últimos 20 años con muy buenos resultados en el servicio de Cirugía del Hospital de Tacuarembó.

Catheterization and MoCaT techniques: What is new?  
Okan Akhan, MD, Prof. (Turkey)

Abstract is not available.

## **SCIENTIFIC ORAL PRESENTATIONS**

Algerian experience in medico-surgical management of hydatid disease since the  
World Congress of WAE in Algeria in 2017

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In Algeria, Echinococcosis is a public health problem. Until 2017, the only therapeutic option for the patients in Algeria was surgery, while international recommendations recommend the addition of medical treatment (antiparasitic) to prevent recurrence. It is in this sense that the Algerian Society of Hydatid Echinococcosis (SAEH) has worked for the establishment of a control programmes to ensure access to quality care for all patients in the country.

This program has two important axes:

-The first is to provide the patient with nationally manufactured antiparasitic medical treatment, which was done in 2018

- secondly, the situation had to be analyzed as a starting point for future actions. This analysis focuses on the collection of data, based on data sheets (pharmaceutical, surgical and medical) prepared by national and international experts and filled by health professionals who care for patients in the different parts of the country. All this will allow us to build a basic surveillance network of all patients who are being cared for. In this communication, we will expose you the work of the group of SAEH since the last World Congress in 2017, and share with you what we are advocating for the future in order to improve the management of this disease in Algeria.

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## Symposium 6

# Diagnosis and classification of human Alveolar Echinococcosis

Clinical and laboratory diagnosis of Alveolar Echinococcosis  
Beat Müllhaupt, Prof. (Switzerland)

Abstract is not available.

Imaging classification of Alveolar Echinococcosis  
Tilman Graeter (Germany)

Abstract is not available.

### SCIENTIFIC ORAL PRESENTATIONS

Evaluation of remnant liver function in patients with hepatic alveolar echinococcosis (HAE) using energy spectrum CT  
Wang Bei, Wenya Liu

**Purpose:** To investigate the correlation between the changes of iodine concentration and the Child-pugh grading score in patients with hepatic alveolar echinococcosis (HAE). **Methods:** 31 patients with clinically diagnosed HAE were included in study. According to the Child-pugh score of their liver function, all the patients were divided into group A 20 cases and group B 11 cases. Their liver function scores were also recorded. Another 15 cases with no liver disease and need for abdominal CT examination were treated as a control group. All patients underwent three-phase CT scan on a HD 750 scanner. The energy CT images of water and iodine - based materials were reconstructed by energy spectrum analysis software. The levels of iodine in the liver parenchyma and the average iodine content in the abdominal aorta and portal vein were measured in the iodine-based images, and the normalized iodine content in the hepatic arterial phase and the portal phase were calculated. the results were analyzed by single factor analysis of variance. The correlation between iodine content and Child-pugh grading score in hepatic arterial phase and portal phase was analyzed by spearman method. **Results:** The levels of iodine in the liver parenchyma of the study group and the control group in the portal phase were higher than those in the arterial phase. The levels of iodine in the arterial phase and the portal phase were higher in the control group than in the control group. The difference of iodine content between the study group and the control group was with statistical significant, and the difference between group A and group B was with statistical significant. The results of spearman analysis showed that the iodine content in hepatic artery and hepatic portal phase was negatively correlated with the Child-pugh grading score. **Conclusion:** The changes of iodine content in patients with HAE was associated with the Child-pugh grading score, and could be used as a supplemented method to evaluate the liver function status.

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AE hepatic lesions: correlation between calcifications at CT and FDG-PET/CT metabolic activity

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Purpose : To correlate the presence of calcifications in alveolar echinococcosis (AE) hepatic lesions to the metabolic activity in 18 FDG PET/CT. Methods : 61 patients were included. Images of FDG-PET/CT were interpreted by two independent nuclear medicine physicians. AE hepatic lesions were classify....ABSTRACT WAS ACCIDENTALLY CUT, IT WAS INCOMPLETE.

Gd-EOB-DTPA-enhanced MRI to evaluate the lesion and liver function in Hepatic Alveolar Echinococcosis

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**Objective** Qinghai is a high incidence area of hepatic alveolar echinococcosis (HAE). Histologically, HAE has an invasive growth characteristic similar to malignant tumors, so we discusses the advantages at hepatobiliary phase of Gd-EOB-DTPA-enhanced MRI, the growth and infiltration characteristics of HAE lesion, and strengthen the clear diagnosis of this disease. **Methods** 44 cases were all scanned with 3.0T Gd-EOB-DTPA-enhanced MRI, and relevant dates of laboratory inspections were collected. **Result** In 44 cases, 12 cases (27.3%) were male and 32 cases (72.7%) were female. The mean age was 42 years old (range: 21 -66 years old). Fifty-three lesions were detected in 44 patients, including 27 cases of parenchymal types, 9 cases of liquefactive necrosis, 9 cases of mixed types, and 8 cases of multiple nodules. The width of infiltration zone at the edge at Hepatobiliary phase and DWI ( $b=0\text{mm/s}^2$ ,  $b=800\text{mm/s}^2$ ) were  $(0.93\pm 0.16)\text{cm}$ ,  $(0.73\pm 0.11)\text{cm}$ , which indicated significant differences between two groups ( $P<0.05$ ). At the hepatobiliary phase, 41 (77.4%) lesions showed better than the portal phase. About 6 (75%) cases named multiple nodules which have small lesions showed more clearer at the hepatobiliary phase than at the portal phase. Furthermore, 44 cases were divided into 3 groups according to the child-pugh classification, including 25 case in group A, 12 cases in group B and 7 cases in group C. Then we found that the degree of clear presentation of lesions was child A (88%, 22/25) > child B (58.3%, 7/12) > child C (42.9%, 3/7) of liver function. **Conclusion** 1. In the hepatobiliary phase, lesions can be shown more clearly, especially the small lesions within multiple nodules, providing the possibility of complete resection of all lesions. 2. The hepatobiliary phase is more advantageous in showing the infiltration zone at the edge of the lesion, providing a more definite resection range for the surgery. 3. Liver function can be assessed indirectly at the hepatobiliary phase.

**Key words:** Gd-EOB-DTPA; Magnetic resonance imaging; Hepatic alveolar echinococcosis; liver function

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Early serological diagnosis of human alveolar echinococcosis  
Philipp KRONENBERG <sup>1/2</sup>, Felix GRIMM<sup>1</sup>, Saša ŠTEFANIĆ<sup>1</sup>, Beat MÜLLHAUPT<sup>3</sup>, Cordula MEYER ZU SCHWABEDISSEN<sup>3</sup>, Sezdbek AITBAEV<sup>4</sup>, Kubanychbek ABDYKERIMOV<sup>5</sup>, Gulnara MINBAEVA<sup>6</sup>, Bruno GOTTSTEIN<sup>7</sup>, Paul TORGERSON<sup>5</sup>, Norbert MÜLLER<sup>7</sup>, Peter DEPLAZES<sup>1</sup>

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**Introduction:** Early serological diagnosis of alveolar echinococcosis (AE) is challenging. Previous studies in Kyrgyzstan and China have documented low sensitivities of different diagnostic tests, regarding patients in early stages of the disease. In this study, we aim to evaluate various diagnostic tests for the early diagnosis of AE. **Material/Methods:** We included 60 confirmed AE patients from Switzerland and 40 from Kyrgyzstan, respectively. The diagnosis was performed via PCR or immunohistochemistry after surgery. All lesions were PNM-staged, clinical data and lesion size were included. Sensitivity and specificity were calculated based on 68 blood donors from both countries and 38 patients with non-parasitic liver diseases. Possible cross-reactions were analyzed, using serum samples of 64 confirmed cystic echinococcosis patients and 144 patients with other parasitic infections. We evaluated 8 native antigens of *E. multilocularis*, 3 of *E. granulosus*, 4 recombinant antigens, and 4 commercially available assays of EuroImmune (Western blot, IFAT, ELISA) and Vector-Best (ELISA, frequently used in Kyrgyzstan). All native antigens of *E. multilocularis* were produced in duplicate with a European and a Kyrgyz isolate (European and Asian haplotype). **Results:** Our test-panel revealed high sensitivities (90-97%) and specificities (95-99%) in most of the tests used for AE patients from Switzerland, regardless of the lesion size. Serological diagnosis of Kyrgyz AE patients achieved sensitivities between 63-93% and specificities between 78-99%. While native antigens were most sensitive to detect Swiss AE infections, recombinant antigen EM95 seems a suitable candidate for the diagnosis of AE in Kyrgyzstan (Sensitivity 85%, Specificity 95%). **Discussion:** Whereas a sensitive and specific early diagnosis of Swiss AE infections is feasible, serological diagnosis of Kyrgyz AE patients remains challenging. We are now evaluating the factors that could explain the differences in test performances between Swiss and Kyrgyz AE patients, considering clinical presentation, age of patients, size of lesions and variations in host-parasite interactions.

**key words:** alveolar echinococcosis, serology, early diagnosis

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## Symposium 7

# Anti-parasitic drug treatment of Human Cystic Echinococcosis

Antiparasitic drug treatment of Cystic Echinococcosis: the evidence of cyst stage specific efficacy

Marija Stojkovic, MD (Germany)

Abstract is not available.

Global access to Benzimidazoles: preliminary results of an ongoing survey

Leonardo Uchiumi, MD (Argentina)

Abstract is not available.

Interactive case series: Would you treat this patient with albendazole?

Marija Stojkovic MD (Germany), Leonardo Uchiumi MD (Argentina), Thomas Junghanss MD (Germany)

Abstract is not available.

### SCIENTIFIC ORAL PRESENTATION

PATENT ON SALTS HAVING BENZIMIDAZOLIC COMPOUNDS: FOCUS ON SODIUM SALT OF RICOBENDAZOLE (Na-RBZ) AND ITS ENANTIOMERS

Gulay VURAL, Mehmet YARDIMCI, Melek KOCAK, Tuba O Yr ERKAL, Ali KURT, Ismail S HAREM, Simone CARRADORI, Ilaria SCIAMANNA, Mar SILES-LUCAS, Massimo FABIANI, Eylem A OZTURK, Andrew HEMPHILL, Britta LUNDSTRÖM-STADELMANN, Roberto CIRILLI, Adriano CASULLI

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**INTRODUCTION:** Among the management options for human cystic echinococcosis (CE), drug treatment is currently based on the use of poorly soluble benzimidazoles (BMZ). However, due to the lack of parasitocidal activity of BMZ, and the requirement for long-term treatment, there is ample room for improvement in terms of efficacy. In this context, new formulations of "Salts of compounds having a benzimidazolic structure" (S-BMZ) was recently patented (PCT/IT2016/000191) for the European and US markets. **MATERIALS AND METHODS:** The invention relates to salts of anthelmintic compounds with a benzimidazolic structure, namely, albendazole (ABZ), Fenbendazole (FBZ), Triclabendazole (TRBZ), or sulphoxides thereof, flubendazole (FLZ), mebendazole (MBZ), oxbendazole (OBZ), thiabendazole (TBZ), cambendazole (CBZ), parbendazole (PBZ) and nocodazole (NCZ). Moreover, the single enantiomers of sulfoxides can be easily produced by HPLC resolution on polysaccharide-based chiral stationary or by asymmetric oxidation. **RESULTS:** Among these S-BMZ, research focused on ricobendazole (RBZ,

albendazole sulphoxide). Dissolution experiments via spectrophotometric measurements indicate that RBZ-Na is highly soluble: 22.83 mg mL<sup>-1</sup> in water vs 0.06 mg mL<sup>-1</sup> of the unsalified form. In addition, the efficacy of RBZ-Na and its enantiomers [(R)-RBZ-Na; (S)-RBZ-Na] has been recently assessed comparatively to conventional ABZ therapy in an experimental mouse model of CE, and demonstrated that mice treated with ABZ-Na, RBZ-Na and (S)-RBZ-Na exhibited statistically significant lower cyst weights than mice treated with conventional ABZ therapy. Histology showed that treatments with RBZ-Na enantiomers and ABZ-Na resulted in degenerated germinal layer morphology and decreased thickness of the laminated layer of the parasite. DISCUSSION/CONCLUSIONS: Because of these encouraging results and the simple synthetic preparation of both racemic and enantiopure forms of S-BMZ, the proposed approach is suitable for a sustainable and affordable large-scale pharmaceutical development of these alkaline salts for the treatment of helminth parasites in humans. This work was supported by the EU FP7, HERACLES project (GA602051).

## Symposium 8

### Treatment of Alveolar Echinococcosis

Is it possible to stop benzimidazole treatment in patients with inoperable alveolar echinococcosis?

Beat Müllhaupt, Professor (Switzerland)

Abstract is not available.

AE at University Hospital of Ulm (UKU). Diagnosis, treatment and follow up data from 280 AE- patients first presentation from 01.01.2011-31.12.2018

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Background: Alveolar echinococcosis (AE) shows a low incidence and controlled clinical studies are hardly feasible. We therefore retrospectively analyzed AE patients with first visit at our center. Material and methods: All patients referred to our center between 01.01.2011 and 31.12.2018 (n=280) were analyzed with regard to clinical presentation,

disease management and outcome. Results: Throughout the last years we observed a strong increase in patients admitted to our center (2011: n=21; 2018: n=43). Median age at diagnosis was 55 years (range 11–89 years). According to WHO-IWGE n=157 (59%) patients had „confirmed“ and n=99 (37%) had „probable“ diagnosis of AE. For 31% (n=78) the finding was incidental and 30% (n=74) did not present any specific symptoms. Curative resection of AE lesions was aspired in n=102 patients, with R0 in 16%, R1 in 8%, R2 in 2%, and Rx in 7% of cases reached. According to the WHO guidelines, benzimidazole therapy (BMZT) for  $\geq 2$  years after resection was recommended. Cases without resection (n=166) received continuous BMZT. BMZT related toxicity was observed in 18% (n=44) of patients. Patients classified as „possible“ (4%) did not receive therapy, but regular follow-up. We considered 24% (n=55) of the patients cured (no relapse > 2 years after BMZT, following resection), or prospectively cured (follow-up < 2 years after BMZT, following resection). The majority of patients with 57% (n=131) has been assessed as chronically stable, n=5 (2%) progressed despite BMZT and one patient died. Notably, AE diagnosis was frequently found in patients with immunosuppression and malignomas, accounting for 20% of AE patients. Conclusions: Increasing patient numbers mirror rising disease awareness and infection rates. AE mostly requires long-term BMZT. Regarding rising incidences and limited curative options, improved prevention and screening is needed. High AE incidence in patients with compromised immune system points towards common factors inducing diminished resistance.

## SCIENTIFIC ORAL PRESENTATIONS

East meets West in pushing the envelope of End-stage Alveolar Echinococcosis  
Surgery: Ex Vivo Liver Resection and Autotransplantation

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Introduction: Radical lesion resection plus medication therapy for alveolar echinococcosis (AE) still remained the best curable option, but not always possible in advanced cases with needing for challenging surgeries even up to liver transplants (LT). *Ex vivo* liver resection and autotransplantation (ELRA) has been encouraging in Chinese end-stage patients. However, western researchers questioned the validity of this procedure in Europe. Whereas, they might have lost the underlying surgical and epidemiological situations among these subjects. Material and Method: Ninety-five end-stage AE cases received ELRA within the last 8.5 years. Specific surgical indications, basic and applied technical aspects, multidisciplinary decision- making process, clinical and individualized case management as well as prognosis were systematically assessed in depth. Besides, transplantation techniques and medication therapy for end-stage AE setting was reviewed comprehensively. Results: ELRA was successful in all patients without intraoperative mortality, and relevant technical aspects were well reported previously. Among this sample, one third of the cases were preoperatively administrated with albendazole but still couldn't prevent disease progress, which was well controlled by radicality through ELRA surgery compared to allogenic LT. Overall mortality was still under 11%, which indicated gradual improvement compared to that from initial years' data, and 100% disease-free survival rate in survivors was obtained. From the perspective of epidemiology of AE and surgical indication for ELRA, China harbors a large amount of advanced cases that were difficult to control as they already become symptomatic with

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end-stage liver disease signs that should be surgically treated with strict indications. Conclusion: What Chinses AE patients encountered now is the situation that the West faced 30 years before, when cases were diagnosed at late stages, for which only albendazole could do nothing much to control disease progress as proved in our sample. Keywords: surgical indication, clinical prognosis, medication therapy.

Is radical surgery still the gold standard for European patients with alveolar echinococcosis?

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Introduction: Anti-parasitic treatment with benzimidazoles has improved survival rates of alveolar echinococcosis (AE) considerably. Radical surgery (RS) is still considered the best therapeutic option; however, no prospective evaluation of the new therapeutic attitudes and their influence on the outcome is available. Material and Method: AE cases from a single center within the French Registry, diagnosed between Jan 1st 2003 and Dec 31st 2011, with treatment assignment at diagnosis by a multidisciplinary team, were included in the study. The 5-years follow-up was analyzed according to 4 groups: G1) RS; G2) no RS: contraindication to surgery or patient's refusal; G3) no RS: technically impossible or too risky; G4) no RS: inactive or very small lesions. Results: Among 85 patients (60% males, median age 64-yrs), 90% were P1 or P2 at PNM staging; 32 patients were in G1; 16 in G2; 31 in G3; 6 in G4; 38% of patients had RS; 30% of G3 patients had no RS despite P1 or P2 stages, because of locations in the 2 liver lobes or size of lesions (higher in G3). There was no difference between groups for global and event-free survival, as well as survival without progression; there was a non-significant trend to lower survival in G2 and G3; however, death was related to associated conditions; only one death, in G3, was related to AE. Progression rate was significantly higher in G2 at the end of follow up, and was associated with non-observance of the anti-parasitic treatment, and with immunosuppression status (more frequent in G2). Discussion / Conclusion: Nearly all European AE patients have a favorable outcome 5 years after diagnosis, whatever the initial treatment assignment. Associated conditions are the main causes of death. Currently, the PNM staging is not a good predictor for treatment assignment. Better staging scores and influence of the multidisciplinary decision on the therapeutic strategy deserve to be studied.

Key words: alveolar echinococcosis, care management, follow-up, treatment, surgery

To predict the correlation between calcification and its biological activity of Hepatic alveolar echinococcosis

Ye Shuai Liu Wen-ya, Liu Wen-ya

OBJECTIVE: To investigate the relationship between different types of calcification and its biological activities in the clinical application. METHODS: Retrospective analysis of 60 cases of hepatic hydatid disease image results, based on the CT findings of calcification

different imaging characteristics of the classification, the same period of PET-CT examination to measure the SUVmax. The correlation analysis was carried out by using rank and test Kruskal - WallisH by different typing SUVmax. Results: 60 cases of patients with hepatic *alveolar echinococcosis* were divided into A, B and C by different calcifications. The median SUVmax of A, B and C were 3.41(2.17-3.75), 7.45(6.77-9.01) and 6.67(6.28-9.01), respectively. The median SUVmax was statistically significant ( $\chi^2=5.429, P < 0.05$ ). Conclusion: There is some correlation between the imaging types of calcification of hepatic alveolar echinococcosis and its biological activity.

Immune exhaustion of T cells in alveolar echinococcosis patients and its reversal by blocking checkpoint receptor TIGIT in a murine model

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The cestode *Echinococcus multilocularis* (*E. multilocularis*) infection is a serious health problem worldwide, which causes alveolar echinococcosis (AE), a tumor-like disease predominantly located in the liver, and able to spread to any organs. Until now, there have been few studies to explore how T cell exhaustion contributes to parasite's escape from immune attack and how it might be reversed. In this study, we found that liver TIGIT expression was significantly enhanced, and positively correlated with lesion activity in AE patients. High TIGIT expression on both liver-infiltrating and blood T cells was associated with their functional exhaustion, and its ligand CD155 was highly expressed by hepatocytes surrounding the infiltrating lymphocytes. In co-culture experiments using T cells and hepatocytes, CD155 induced functional impairment of TIGIT<sup>+</sup> T cells, and in vitro blockade with TIGIT antibody restored the function of AE patients' T cells. Similar TIGIT-related functional exhaustion of hepatic T cells, and an abundant CD155 expression on hepatocytes were observed in *E. multilocularis*-infected mice. Importantly, in vivo blocking TIGIT prevented T cells exhaustion and inhibited disease progression in *E. multilocularis* mice. Mechanistically, CD4<sup>+</sup> T cells were totally and CD8<sup>+</sup> T cells partially required for anti-TIGIT-induced regression of parasite growth in mice. Conclusion: This study demonstrates that *E. multilocularis* can induce T cell exhaustion via inhibitory receptor TIGIT, and that blocking this checkpoint may reverse the functional impairment of T cells, and represent a possible approach to *E. multilocularis* immunotherapy.

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## ROUND TABLE 3

### Countries with control / elimination programs of Cystic Echinococcosis

#### CONTROL / ELIMINATION PROGRAMME OF CYSTIC ECHINOCOCCOSIS IN TURKEY

Nazmiye Altintas, PhD (Turkey)

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President of Turkish Association of Hydatidology (TAH)

Cystic echinococcosis (CE) caused by *E. granulosus* and alveolar echinococcosis (AE) caused by *E. multilocularis* have been found in Turkey. Cystic echinococcosis occurs throughout Turkey, whereas AE predominantly occurs in the Eastern Anatolia region. CE is still one of the most important helminthic diseases in Turkey.

CE are transmitted in domestic lifecycles involving dogs and livestock, especially in regions with extensive livestock husbandry and slaughter carried out at farms without proper meat inspection. That is why, although CE has been recognized in Turkey since 1861, a consistent state policy for disease prevention and control has not been developed yet and CE continues to be a major health problem. The main reason for facing this public threat in Turkey in such a large scope is because this topic has not yet become a priority for the authorities. Exposure of humans to *E. granulosus* is common due to the lifestyle of the people who live in close proximity to sheep and dogs. Most people live in rural areas and are engaged in animal husbandry. The public awareness on this issue is very low, thus people try to protect themselves and their children with hearsay information that are wrong and useless. Another reason is, once a year, a sacrificial festival is held during which a lot of sheep and cattle are slaughtered and also for daily meat requirements. the meat of infected intermediate hosts is commonly fed to stray dogs., with a high proportion of uncontrolled slaughter which facilitates transmission of the parasite.

Although the present number of stray dogs and owned dogs is unknown in Turkey, according to some of the local studies, *E. granulosus* infection in dogs ranges between 0.32% and 40%. The prevalence of CE in domestic animals ranges from 3.5% to 58.6% has varied widely with geographical location. In humans, Altintas (2003) reported that the estimated surgical cases rates ranged from 0.87 to 6.6 per 100,000 inhabitants between 1987 and 1994. Still numerous human cases are reported regularly from medical centres in different parts of the country.

Control programme development has been started to discuss with the authorities from Ministry of Health and Ministry of Agriculture after establishing the Turkish Association of Hydatidology (TAH) in 1999. TAH is the only Non Governmental Organization (NGO) in Turkey that works on "cystic echinococcosis". Then CE became a "notifiable disease" in 2005 by Ministry of Health. But still there is no accurate information about the disease because of the surveillance system. So, CE urgently needs attention both for protecting public health and animal welfare in Turkey.

Keywords: Cystic echinococcosis, *Echinococcus granulosus*, control programme, Turkey

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Key considerations on implementation, monitoring, evaluation of echinococcosis  
control actions  
Marshall Lightowlers, PhD (Australia)

Abstract is not available.

Control programme Kyrgyzstan  
Paul Torgerson, PhD (Switzerland)

Abstract is not available.

Large-scale Vaccination of EG95 Vaccine Significantly Interfered with the Dog-Sheep  
and Goat Transmission Chain of Hydatidosis  
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Cystic Echinococcosis (CE) also known as Hydatidosis caused by the larvae of *Echinococcus granulosus* is endemic and important zoonosis in the pastoral and semi-pastoral area of North-Western China. Since 2010, the government conducted the control measures using praziquantel in dogs in the key endemic areas, which has reduced the transmission of the disease to intermediate hosts, but the epidemic situation remains serious. Pilot field trials of the recombinant hydatid subunit vaccine (EG95) in sheep and goat herds have been completed in three provinces, and the results were very satisfactory. In 2016, the central government launched comprehensive compulsory vaccination of sheep and goats with EG95 vaccine in Xinjiang, Sichuan, Qinghai, Gansu, Tibet, Ningxia and Inner Mongolia, that is, newborn lambs received two vaccinations, 4-week apart, and a single booster one year later. To date, 40.91 million, 47.76 million and 44.92 million doses of EG95 vaccine were used in these seven provinces in 2016, 2017 and 2018, respectively. The immune density and positive rate of anti-EG95 antibodies was 61.20% -94.90% and 60.09% -89.20%, respectively. Slaughter examination and necropsy of sheep and goats revealed that the prevalence has decreased from 5.34%-44.72% in 2014/2015 to 1.23%-20.16% in 2017/2018. The prevalence of *Echinococcus granulosus* in dog population has decreased from 3.12%-29.63% to 1.04%-13.80% determined by arecoline purgation, this number has decreased from 12.88% to 2.04 % in Ningxia when coproELISA was used. After three years of comprehensive immunization in these difficult and remote environments, the prevalence decreased significantly in both intermediate host animals and dogs, demonstrating excellent clinical immune efficacy of EG 95 vaccine, indicating that the vaccination has effectively interfered with the dog-sheep and dog-goat transmission chain, which provides an economical, efficient and practical tool for controlling hydatidosis.

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## SCIENTIFIC SESSION 1 (oral presentations)

Records of deaths attributed to Echinococcosis in Brazil: 1995-2016

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**Introduction:** We present records of deaths attributed to echinococcosis in Brazil in the period 1995 to 2016. Previous reports of echinococcosis deaths did not have national coverage, were informed by individual registries or covered short periods. Given the likely underreporting of the condition, analyses of multiple surveillance streams are warranted. **Materials and Methods:** We retrieved data from two national registries, the Hospital Information System (HIS) and the Mortality Information System (MIS), to describe the most relevant demographic, epidemiological, and healthcare related features of deaths attributed to echinococcosis in the period of study in Brazil. **Results:** The HIS registered 185 deaths, out of 7,955 hospital recorded cases of echinococcosis, and the MIS 113 deaths. All states in Brazil recorded deaths, most of them (>40%) in patients over 45 years old. The HIS showed large variability in case fatality rates across states, possibly indicating some underlying heterogeneity in the diagnosis and quality of healthcare. **Discussion:** Our results only capture the most severe forms of the disease and hence they do not inform the overall burden of echinococcosis in Brazil. The current registration of deaths attributed to echinococcosis is not free from problems. For example, double counting is likely to occur between the two surveillance sources although the distribution of records throughout the period across the two databases was rather asynchronous. In addition, coding changes in the HIS in the late 90s saw the integration of echinococcosis cases with those of other pathologies and led to the loss of record specificity. This is seen in the drastic decrease of registered echinococcosis deaths in subsequent years. Our results show the wide geographical distribution of deaths due to echinococcosis, and highlight the need to expand the notification of the disease across the country, currently only in place in the state of Rio Grande do Sul. **Keywords:** Brazil, deaths, surveillance sources.

**Mathematical modelling of Echinococcus granulosus to identify optimal intervention strategies: a case study from five pilot interventions in the Peruvian highlands**

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**Introduction:** Cystic Echinococcosis (CE) in Peru is still a major health problem with over 20,000 human cases reported between 2009 and 2014. In 2015, health authorities initiated 5-year pilot projects to assess the effectiveness of a number of control measures, in 5 different regions in the highlands. Those measures include dog deworming, sheep vaccination, and anthelmintic treatment of sheep, as well as combinations of these. Using data from the pilots, we will present a calibrated mathematical transmission model that

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can help inform control strategies. Material and methods: We have adapted and extended an existing compartmental, deterministic, prevalence-based transmission model to simulate *Echinococcus granulosus* transmission in Peru, with the aim of simulating the various intervention strategies tested in the pilot control projects. The model was ran for multiple settings and forecasts in prevalence reduction were obtained for the alternative control options. The model is planned to be fitted to the copro-ELISA data collected during the 5-year pilot. Results: Our preliminary model results include: i) a sensitivity analysis of model parameters that are setting-specific and capture the range of prevalence observed in the South American region, and ii) simulations of the impact of the control measures implemented in the pilot projects. The model validation plans will be also presented. Discussion: We will discuss how the transmission model will be linked to an economic evaluation, based on dog prevalence outcomes of the different pilot interventions. Furthermore, we will explore how the modelling framework will be used to assess the impact of interventions on CE human health burden, and the scaling-up of interventions to support a national control strategy for Peru. In the longer-term, the project aims to inform the regional action plan for CE control.

Epidemiological Surveillance of Human Hydatidosis between the years 2018 at the beginning of the year 2019 in the Ayacucho's Region - Perú.  
Huamani-Basilio Nelly N, Sulca Guisela L, Cruz Miguel A.  
Ayacucho's Health Regional Direction

Hydatidosis is a disease caused by the cestode *Echinococcus granulosus* which is an important problem for public health and affect patient's economy, whose factors associated are diverse, however, these vary according to the region where it is presented; so that, in the present study we determined the characteristics of epidemiological surveillance of Human Hydatidosis in the region of Ayacucho, between 2018 and 2019 years. The methodology consisted of a descriptive analytical design, that data were obtained from 215 notifications of Notiweb from the region of Ayacucho; the following variables of interest were: Health establishment, laboratory tests, age, sex, origin and altitude. The data was ordered with Microsoft Excel software, and the results were 146 confirmed cases, 56 discarded cases and 13 presumptive cases. Also, determined 20 health facilities, of which 50% represent the Ayacucho's Hospital, being the center with highest resolution in the region, presenting from 2 years to 89 years, which corresponds to the age group over 60 years with 19%, and between 18 to 29 years with 18%; with respect to sex, 66% affect the female population and 34% at male population; regarding provenance, 16% of the cases confirmed come from the district of Santiago de Lucanamarca, and 11% from the district of Sancos; according to the altitude, are presented from 2,535 m.s.n.m in the district of Pacaycasa and 3,502 m.s.n.m. in the district of Querobamba. Therefore, it is concluded that the results of the Epidemiological Surveillance of Human Hydatidosis demonstrate the presence of the disease in the Ayacucho's Region, with the highest cases registered in the districts of Santiago de Lucanamarca and Sancos.

Establishment and Innovative Practice of Integration System of Prevention, Diagnosis & Management (ISPDM) for Human Echinococcosis

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Echinococcosis is a neglected tropical zoonotic disease caused by the larva stage of *echinococcus* family and distributed globally. China harbors the severest disease burden regarding to two main types of echinococcosis (cystic echinococcosis/CE; alveolar echinococcosis/AE), threatening public health and posing a heavy economic burden. And, western China bears over 90% of world AE cases alone. Great achievements have been made in controlling the disease since the foundation of New China: rate of clinical cure has improved while significantly reducing the mortality and infection ratio of both human and animal subjects. Especially from the 21<sup>st</sup> century, our state has obtained encouraging advances in echinococcosis genetics, genomics, molecular epidemiology, immunology, diagnostic tools, treatment approaches, prevention strategies, vaccine development. Then after, the establishment and innovative practice of Integration System of Prevention, Diagnosis & Management Integration (ISPDM) for human echinococcosis has gained substantial progress and achieved high socio-economic benefits.

**KEYWORDS** echinococcosis, scientific prevention, definitive diagnosis, innovative surgery, medical treatment

#### Educational tools for human cystic echinococcosis prevention among school students on an endemic area of Peru

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Human cystic echinococcosis (HCE) in an endemic disease in Peru. The ignorance about this disease and unsafe practices expose people for be infected, and children are the most vulnerable. Therefore, the aim of the study was to validate four educational tools for HCE prevention among third to fifth grade students from a basic education school in Junin, Peru. The quasi-experimental study considered three phases: Phase 1) Design of educational tools, Phase 2) Expert validation of tools, Phase 3) Field validation of tools. To contrast the knowledge before and after the intervention, a ten-item test was applied. Four educational tools were generated: a color paint storybook, a board game, a flipchart and a museum. The expert validation determined a strong validity for the flipchart and the storybook (Aiken V=1.14 and 1.21, respectively), and an acceptable validity for the board game and the museum (Aiken V=1.08 and 1.03, respectively). On the field validation 131 students participated, 42.8% were women and 57.2% men, with ages between 8 and 13 years. Most students modified their knowledge, the number of correct answers between the pre and post test were different ( $p < 0.05$ ), proving that the educational tools promote the acquisition of knowledge for HCE prevention. The intervention included the topics on school classes without altered the Peru National Basic Education Curriculum. We expect the knowledge lasts, and through the time favors the change of attitudes and practices for reduce the risk of HCE infection.

**Keywords:** Hydatidosis, health education, zoonoses, health promotion.

Effectiveness of ten years of control of *E.multilocularis* by deworming owned dogs using praziquantel in a highly endemic area of Tibetan communities, China  
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**Introduction:** Human alveolar echinococcosis (AE) was found as an emerging public issue in Tibetan communities of Shiqu county just 20 years ago. On St. Lawrence Island, Alaska, transmission of *E. multilocularis* was controlled by regular deworming of owned dogs over a 10-year period since the 1970s. In Tibetan communities of the Tibetan Plateau, with a continental setting, the control of transmission is a great challenge due to complex epidemiology, disease ecology, geography and socio-cultural factors. However, a control programme to deworm owned dogs has been carried out since 2006. **Objective:** to understand the change in infection of *E.multilocularis* in dogs after ten years of deworming of owned dogs using praziquantel. **Material and Method:** The study was carried out in all accessible 17 townships in Shiqu County of China in October of 2016. Owned dogs were purged using arecoline hydrobromide to collect fecal samples, from which tapeworms were collected and subjected for PCR analysis. Small mammals were trapped to collect viscera; kidney were subject for PCR identification of species of the small mammal; lesions were subject for PCR identification of *E. multilocularis* infection. Fisher exact test were used to understand the difference between prevalences. **Results:** the *E. multilocularis* infection rate in dogs was significantly reduced from 7.23%(25/346) to 0.55%(1/181) in 3 townships. The overall infection rate was 1.44% (14/970) in dogs from 17 townships. The prevalence rate of *E. multilocularis* infection was 1.56% (3/192) and 11.59% (8/69) for *Ochotona curzoniae* and *Neodon fuscus* respectively. **Discussion/Conclusion:**The control programme was effective in reducing the *E. multilocularis* infection in dogs, but did not produce impact on the infection in small mammals. One possible reason might be the programme did not eliminate the stray dogs, which actively disseminate the eggs of *E. multilocularis*., and also did not concern wildlife definitive hosts such as the red fox and the Tibetan fox, both present in the area.

## SCIENTIFIC SESSION 2 (oral presentations)

In vitro and in vivo efficacy of DNA damage repair inhibitor Veliparib combined with artesunate against *Echinococcus granulosus*

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**Objective:** In this study, we evaluate the effects of the DNA damage repair inhibitor Veliparib combined with artesunate (AS) as a new formulation on CE both in vitro and in vivo. **Methods:** in vitro assay, protoscoleces (PSCs) of *E. granulosus* were incubated with ABZ, Veliparib, AS-L/M/H, AS-L/M/H+Veliparib, respectively. After drugs incubation, we used eosin staining and alkaline phosphatase activity assay to detect PSCs activity; transmission electron microscopy (TEM) to observe the ultrastructural change of PSCs; comet assay, immunofluorescence, and qRT-PCR assay to evaluate the DNA damage, 8-hydroxydeoxyguanosine (8-OHdG) and RPS9 gene expression, respectively. In vivo assay, model mice infected with CE intragastric administrated with ABZ, AS-L/H and AS-H+Veliparib for 6 weeks. During administration, B-ultrasonography monitor the volume change of vesicles once a week. After administration, calculate the cysts inhibition rate, observe the ultrastructural changes of vesicles and detect the 8-OHdG expression. **Results:** The maximum protoscolicidal effect was found with AS-H+Veliparib, and ultrastructural revealed that germinal layer (GL) cells were reduced, and lipid droplets appeared. Comet assay, immunofluorescence and qRT-PCR results revealed that DNA damage, cometary olive tail moment (OTM) value and the expression of 8-OHdG increased significantly in AS-H+Veliparib group, and the expression of RPS9 gene in AS-H+Veliparib group was down-regulated (versus AS-H). B-ultrasonography showed that maximum potentiation was seen at day 15 in AS-H+Veliparib, and led to reductions of 55.40% in cysts weight compared with the model ( $P<0.01$ ), which was better than AS-H (52.84%) and ABZ (55.35%). Ultrastructural of cysts in AS-H+Veliparib revealed that the GL damage, the microtriches disappeared. The vesicles GL shows the expression of 8-OHdG. **Conclusion:** Our study revealed that AS alone or AS combined with Veliparib both have anti-CE effect in vitro and in vivo, especially the combination group. The study initially proved AS combined with Veliparib might be the promising drug candidates in anti-CE chemotherapy.

**Keyword:** Cystic echinococcosis, Artesunate, Veliparib

**Effect of siRNA interference targeting EgRPS9 gene on DNA oxidative damage of *Echinococcus granulosus* protoscoleces**

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**Objective** To investigate the effect of siRNA interference with EgRPS9 gene expression on DNA oxidative damage in protoscoleces of *Echinococcus granulosus* by electroporation. **Methods** EgRPS9-siRNA and scrambled siRNA were transfected into *Echinococcus granulosus* protoscoleces by electroporation. It included EgRPS9-siRNA-184, 337, and 448 interference groups, H<sub>2</sub>O<sub>2</sub> intervention, scrambled control and blank control groups. Three days after electroporation, the protoscoleces were treated with the maximum tolerated concentration of H<sub>2</sub>O<sub>2</sub> for 1 h, stained by eosin, and observed under an inverted fluorescence microscope to calculate the survival rate of protoscoleces. The cultural medium in each group was collected for alkaline phosphatase activity assessment. RNA was extracted from protoscoleces using Trizol, and EgRPS9 mRNA expression was assessed by qRT-PCR. The single cell gel electrophoresis (SCGE) was used to detect DNA damage before and after transfection.

The content of reactive oxygen species (ROS) in protoscolecemes was assessed using and ROS detection kit. Data were analyzed using the SPSS 22.0 software. Results The maximum tolerated concentration of H<sub>2</sub>O<sub>2</sub> for *Echinococcus granulosus* protoscolecemes was 0.4 mM, and the intervention duration was 1 h. The siRNA sequences were effectively transfected by electroporation. Green fluorescent spots were clearly seen in the interference groups and scrambled control group, while none appeared in H<sub>2</sub>O<sub>2</sub> intervention and blank control groups. The survival rates of protoscolecemes in the EgRPS9-siRNA-184, 337, and 448 interference groups were (59.45±5.25)%, (58.82±4.35)% and (26.59±2.76)%, respectively. Thus the EgRPS9-siRNA-448 was considered as the most effectively interfering sequence. The alkaline phosphatase activity in the EgRPS9-siRNA-184, 337, and 448 interference groups were 0.041±0.004, 0.038±0.003 and 0.008±0.001, respectively, with significant difference between EgRPS9-siRNA-448 and H<sub>2</sub>O<sub>2</sub> intervention group (0.091±0.001) ( $P < 0.01$ ), which further supported the highest effectiveness of EgRPS9-siRNA-448. The EgRPS9 mRNA expression levels in the EgRPS9-siRNA-184, 337, and 448 interference groups were 0.623±0.074, 0.599±0.041 and 0.223±0.060, respectively, with significant difference between EgRPS9-siRNA-448 and H<sub>2</sub>O<sub>2</sub> intervention group (1.501±0.022) ( $P < 0.01$ ), again verifying the highest effectiveness of EgRPS9-siRNA-448. Under the same electrophoresis condition, the DNA fragments in the EgRPS9-siRNA-448 group migrated from the nucleus to the anode, with a smearing tail. The Olive tail moment in the EgRPS9-siRNA-448 group was 14.357±2.005, difference from the H<sub>2</sub>O<sub>2</sub> intervention group (6.449±1.510) ( $P < 0.05$ ). The ROS content in the EgRPS9-siRNA-448 group was 16.105±0.247, difference from the H<sub>2</sub>O<sub>2</sub> intervention group (7.265±0.103) ( $P < 0.05$ ). Conclusion The EgRPS9 gene plays an important role in DNA oxidative damage. The EgRPS9-siRNA-448 can specifically interfere with the expression of EgRPS9 in *Echinococcus granulosus* protoscolecemes, and reduce the ability of DNA oxidative damage repair in protoscolecemes.

Key words: EgRPS9; siRNA; *Echinococcus granulosus* protoscolecemes; DNA oxidative damage

#### Multiple haplotypes of *Echinococcus granulosus sensu stricto* in single naturally infected intermediate hosts

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Cystic echinococcosis is a disease that affects both humans and animals, caused by cryptic species complex belonging to the platyhelminth *Echinococcus granulosus sensu lato* (*s.l.*). This disease is distributed worldwide, with *E. granulosus sensu stricto* (*s.s.*) being the most widespread of the species. High genetic variability has been demonstrated within *E. granulosus s.s.* studying single cysts per infected animal identifying a number of different haplotypes. However, few studies have addressed the genetic diversity of this parasite within a single intermediate host with multiple *Echinococcus* cysts. Also, it remains unknown if specific haplotypes of *E. granulosus s.s.* produce differences in biological features of the cyst. Here, we use the full length of the mitochondrial gene *cox1* to determine *E. granulosus s.s.* haplotypes in samples from both cattle and sheep which harboured more than one cyst in different areas in Chile, where this parasite is endemic. We found 16 different haplotypes in 66 echinococcal cysts from 10 animals, and both cattle and sheep can harbour up to five different haplotypes of *E.*

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*granulosus s.s.* in the same animal. Regarding cyst fertility, five animals had both fertile and infertile *Echinococcus* cysts in both single and multiple haplotype infections. There was no association between haplotype and cyst fertility, size or adventitial layer characteristics. Sampling and sequencing every *Echinococcus* cyst found in the intermediate host reveals a high molecular variability. We speculate that multiple haplotype infections could also suggest that intermediate hosts come from hyperendemic areas. Funding: FONDECYT 1190817

Keywords: multiple haplotypes, adventitial layer histology, hydatid cyst fertility

Presence of *Echinococcus multilocularis* in eastern Europe: EmsB microsatellite analyses revealed main influence of European historical focus

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Using the EmsB microsatellite, the spatial and temporal spread of *E. multilocularis* in western Europe has been described as resulting from a mainland-island system with transmission across the historical focus and peripheral areas. Nevertheless, very limited data are available from eastern European countries when it would be necessary to understand the history of expansion. In this aim, the genotyping of 546 *E. multilocularis* samples from Czech Republic (26 samples), Slovakia (260), Hungary (83), Serbia (5), Estonia (89), Latvia (42), Ukraine (11), Russia (23) and Turkey (7) resulted to the description of 55 EmsB profiles. 41 profiles correspond to European genotypes of which 35 were already identified in Europe according to comparison with the collection of 1627 samples previously genotyped. The two profiles the most represented are also those that have obtained the most correspondence with the collection samples. Samples from Czech Republic and Slovakia generally clustered together as those from Estonia, Latvia and Ukraine. Samples from Hungary are well distributed among 14 different EmsB profiles including the samples from most of the others countries when samples from Serbia clustered with neighboring countries Slovakia, Hungary and Czech Republic. If two samples from Turkey and one from Russia also grouped with European profiles, the others appeared to clustered with Asian profiles. Asian profiles also included one sample from both Slovakia and Serbia. Furthermore, three samples from Russia are grouped with North American profiles from two different origins: two are close to the samples from Alaska and the other one clustered with those from Svalbard Island. These results are consistent with the expansion from one country to another by the dispersal movement of foxes, which may have been initiated from the historical focus. However, the presence of Asian and North American EmsB profiles highlighted another influences of expansion in eastern Europe.

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## ROUND TABLE 4

# Continental project experiences on Cystic Echinococcosis

CESSARI: Research on Cystic Echinococcosis in Sub-Saharan Africa

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**Introduction:** Cystic echinococcosis is widespread in sub-Saharan Africa, although the presence in some regions is unclear. A considerable amount of research has been conducted over the past five decades particularly in eastern Africa, one of the world's hotspots of human CE. However, even there only few foci are well researched, while the situation in other parts is virtually unknown. The project aimed to bundle local scientific expertise by strengthening research facilities, mobility and communication in order to address open questions concerning presence, frequency and genetic identity of CE in sub-Saharan Africa. **Materials and methods:** From 2009 to 2018, funding was obtained from the German Research Foundation (DFG) for researchers of fourteen medical, veterinary and biological research institutions in Sudan, Ethiopia, Kenya, Uganda, Ghana, Zambia, Namibia, South Africa and Germany as key partners, plus several associated researchers from elsewhere. Six basic molecular laboratories were established or upgraded in Sudan, Kenya, Uganda and Ghana, and four mobile ultrasound units were provided for Sudan, Kenya, Uganda and Zambia. International training courses were organised for molecular biology, ultrasound diagnostics, percutaneous intervention techniques and epidemiology. **Results:** CE prevalence in humans and livestock was established or re-assessed. All five *Echinococcus* spp. that cause CE were found in sub-Saharan Africa. The distribution of the various taxa was very uneven, and prevalence of human CE was correlated with the presence of *E. granulosus sensu stricto* transmitted in a sheep – dog cycle. Lifecycles of most *Echinococcus* species were found to involve wild animals to a previously unknown extent. **Conclusion:** The research initiative has strengthened centers of expertise in African countries and enabled research across national borders. Partly, training activities have shifted from European partners to African institutions, and the scientific output will in future hopefully help to acquire research funding without external assistance.

### THE INTERNATIONAL IMPACT OF HERACLES COLLABORATIVE PROJECT ON CYSTIC ECHINOCOCCOSIS

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**INTRODUCTION:** Cystic Echinococcosis (CE) is one of the most important zoonotic diseases worldwide and was recently assigned to the list of the Neglected Tropical Diseases prioritized by the WHO. Tools for its diagnosis and treatment are currently not

standardized, partly due to the complex and chronic evolution of CE and lack of funding to support prospective multicenter clinical trials, which in turn make data on this infection poorly framed and evidence supported, resulting in yet more neglect. MATERIALS and METHODS: The main goals were to: Identify the population affected by CE in Bulgaria, Romania and Turkey; create the European Register of CE (ERCE); establish a bio-sample repository; set-up and validate new recombinant antigens; identify cyst stage-specific biomarkers; increase drug bioavailability of benzimidazoles. RESULTS: Core achievements are: 1) Creation of the “Heracles Extended Network” with more than 60 centers from 30 countries ([http://www.heracles-fp7.eu/interactive\\_map.html](http://www.heracles-fp7.eu/interactive_map.html)); 2) Biggest research-based cross-sectional ultrasound-based population study (n=24,693) on CE, estimating 151,000 people infected in rural Romania, Bulgaria and Turkey; 3) Creation of the European Register as a prospective case series with more than 2,000 patients registered from 48 centres (<http://www.heracles-fp7.eu/erce.html>); 4) Patent on anti-parasitic soluble drugs: “Salts of compounds having a benzimidazolic structure” (PCT/IT2016/000191); 5) Creation of the Echino-Biobank to sustain experimental and clinical research in this field (n=5,000 samples used); 6) Worldwide collection of human cyst samples for genotyping studies (n=742); 7) First proteomic description of parasite exosomes in echinococcal cyst and identification of biomarker candidates in plasma by quantitative proteomic analysis; 8) Scientific papers published in peer-reviewed journals: 58. DISCUSSION/CONCLUSIONS: The results from HERACLES project (2013-2018) will support governments, international agencies, the European Commission, to harmonize data collection, monitoring and reporting of CE. We see this as breakthrough in the current scenario of CE. This work was supported by the EU FP7, HERACLES project (GA602051; <http://www.Heracles-fp7.eu/>).

#### The state of european register of cystic echinococcosis (ERCE) 5 years after its inception

Patrizia ROSSI, Francesca TAMAROZZI, Fabio GALATI, Okan AKHAN, Carmen M CRETU, Kamena VUTOVA, Mar SILES-LUCAS, Enrico BRUNETTI, Adriano CASULLI, on behalf of the ERCE network\*\*In alphabetical order: Andrea ANGHEBEN, Nikolay BAGMET, Moncef BELHASSEN, Solange BRESSON-HADNI, Fabrizio BRUSCHI, Guido CALLERI, Barbara CASTIGLIONI, Cathy CHEMLA, Leonardo CHIANURA, Balazs DEZSENYI, Maria Teresa GIORDANI, Valbona GJONI, Levan GOGICHAISHVILI, Delia GOLETTI, Majid HARANDI, Gulziya ISMAILOVA, Fazal KARIM, Emma LAPINI, Felix LÖTSCH, Scilla MASTRANDREA, Guido MENOZZI,, André PAUGAM, Alfonso RECORDARE, Malgorzata SULIMA, Antonella TEGGI, Carlo TORTI, Giustina VITALE, Martine WALLON, Lorenzo ZAMMARCHI  
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INTRODUCTION: Cystic echinococcosis (CE) is a neglected zoonosis and the real burden of infection is difficult to evaluate. Given the clinical features of human CE, only a fraction of clinically relevant cases reach medical attention, they are rarely notified and this lack of data contributes to the poor evidence base, which hampers evidence based guidelines for its management. The European Register of CE (ERCE), launched in October 2014 to address the problem is a prospective, observational, multicentre register of patients with CE. Its aims are to gauge the burden of human CE in Europe, bring CE to the attention of health authorities, encourage control policies, and support research on CE. We outline the state and perspectives of ERCE, 5 years after its inception. MATERIALS AND METHODS: The ERCE database was searched (31/03/2019) and data concerning patients' registration, follow-up visits, CE cyst details, and clinical management were analysed.

**RESULTS:** 44 centres in 15 countries contribute to ERCE. Of these, 33 (75%) registered patients and, of these, 18 (54%) recorded at least one visit within the past 18 months. 2,043 patients were registered. 25 (51%) centres recorded follow-up visits after the first patient registration, with a median 25% registered patients having follow-up visits recorded. Cysts characteristics (organ/s involved and/or cyst stage for at least one patient/visit) were recorded by 28 centres. Clinical management details could be analysed for 726 cysts (in 523 patients), for 895 stage-management matched observations. **DISCUSSION/CONCLUSIONS:** ERCE has been constantly expanding, with several Centres contributing data. ERCE has successfully achieved the objective of highlighting the burden of CE in Europe, but number and quality of data need to be improved if all its ambitious goals are to be met.

**Keywords:** cystic echinococcosis, prospective register of cases, epidemiological and clinical surveillance

The South American Initiative for the surveillance, control and prevention of Cystic Echinococcosis/Hidatidosis  
Edmundo Larrieu, PhD (Argentina)

Abstract is not available.

Molecular-epidemiological studies on pathways of transmission and long lasting capacity building to prevent cystic echinococcosis infection: outline of peritas project  
Francesca TAMAROZZI, Gerardo ACOSTA-JAMETT, Edmundo LARRIEU, Saul SANTIVANEZ, Leonatdo UCHIUMI, Maria J PERTEGUER, Mar SILES-LUCAS, Adriano CASULLI

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**INTRODUCTION:** The WHO advocates the control of echinococcosis and general hygiene education is envisaged among control strategies for cystic echinococcosis (CE). Although ingestion of contaminated food/water and direct contact with infected dogs are classically mentioned as infection sources, the precise sources and at-risk behaviours for human infection have not been clearly individuated. Addressing specific at-risk behaviours is expected to increase the effectiveness of hygiene education. PERITAS project aims to elucidate through an integrated, systematic approach the pathways of transmission of *Echinococcus granulosus*. **MATERIALS and METHODS:** The study design is a cross-sectional prevalence study of human infection followed by a case-control molecular epidemiology study on several matrices, comparing level of contamination in high vs low endemic areas. Prevalence of human CE in active stages will be identified by population-based ultrasound surveys in selected areas of Argentina, Chile, and Peru. Sampling of different matrices in these identified areas will be performed, followed by application of molecular techniques to identify contamination and species/genotype of the parasite and an ad-hoc questionnaire focused on specific habits and behaviours will be provided to participants. In addition, biological samples will be collected to feed the biological repository Echino-Biobank, and training and feed of the International Register of CE (IRCE), will allow long-lasting capacity building. **RESULTS:** The analysis of the data obtained in clusters of active human CE infection will allow the identification of matrices

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contaminated by *E. granulosus* eggs and of at-risk behaviours/habits associated with odds of CE infection. **DISCUSSION/CONCLUSIONS:** The direct applicability of project results will allow the implementation of more precisely-targeted human-centred control interventions, saving time and costs of control activities and increasing the efficacy of control programs for the prevention of new human infections. This work is supported by EU-LAC Health project and National funding agencies of the participating institutions - PERITAS project (<http://eulachealth.eu/>).

**Keywords:** cystic echinococcosis, pathways of transmission, environmental contamination

**Wp2\_Molecular/Serological Epidemiological Studies Recombinant Antigens And Serological Tests**  
Mar Siles, PhD (Spain)

Abstract is not available.

**ERANET-LAC PROJECT: DEVELOPMENT OF NEW DIAGNOSTIC AND TREATMENT OPTIONS FOR HELMINTHIC NEGLECTED DISEASES (NDTND)**

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**INTRODUCTION:** Our project aims at developing new therapeutic and diagnostic tools to contribute to the control of neglected diseases caused by helminthic parasites, such as cystic (CE) and alveolar (AE) echinococcosis. **MATERIALS AND METHODS:** Specific *Echinococcus* molecules are being evaluated as new therapeutic/diagnosis targets using bioinformatics, molecular biology, biochemistry and biophysical methods integrated with relevant clinical and epidemiological information. **RESULTS:** The evaluation as therapeutic targets of highly expressed, specific molecules that may exert essential functions for the parasite, such as microRNAs and lipid binding proteins is underway. The small RNAs and proteins actively secreted by larval stages of these parasites are being characterized to assess their potential as biomarkers. Ultrasound surveys and sera collection from humans affected by CE are being performed in order to link the potentially new biomarkers in the stage specific approach according to WHO-IWGE (Informal Working Group on Echinococcosis). **CONCLUSION:** The expected outcomes of the project are the development of new compounds that bind and inhibit essential and unique molecules of these parasites, and to find new detection tools to improve the status of both treatment and early diagnosis of these complex and neglected parasitic diseases.

This work was supported by ERANet-LAC 2<sup>nd</sup> Joint Call (<http://www.eranet-lac.eu>) and the National funding agencies of the participating institutions - NDTND project.

**KEY WORDS:** cystic and alveolar echinococcosis, microRNA, Lipid Binding Proteins.

## SCIENTIFIC ORAL PRESENTATIONS

### ULTRASOUND SCREENINGS IN RIO NEGRO PROVINCE, ARGENTINA: PRELIMINARY RESULTS FROM STAGE 1 OF PERITAS PROJECT

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**INTRODUCTION:** PERITAS project, is aiming to elucidate the pathways of transmission of cystic echinococcosis by two sequential stages to be carried out in Argentina, Chile, and Peru. STAGE 1 is a cross-sectional ultrasound (US)-based prevalence study to identify endemic villages/areas where the following STAGE 2 case-control study will be implemented at household and community level, detecting and comparing matrices contamination with eggs/DNA of *Echinococcus* spp. **MATERIALS AND METHODS:** the selected broad investigation area for project stage 1 in Río Negro Province, Argentina, was Ramos Mexía. The urban and its rural areas (Yaminué and Treneta) were evaluated. Out of 1,214 inhabitants, 581 (48%) were evaluated by abdominal US. 121 inhabitants were 5-14-year-old (20.8%) and 521 (79.2%) were over 15-year-old. 326 inhabitants were females (56%) and 255 males. 451 (77.4%) were from the urban area and 132 from its rural areas. Median age was 36.4 years (range 1 to 82). **RESULTS:** 29 cases (5%) were CE-positive by abdominal US with 38 cysts (34 in the liver, 3 in the spleen and 1 in the kidney). 18 cases (62%) were currently living in the urban area, and 11 (38%) in the rural areas. 23 (3.9%) were new cases. No new cases were found in children. 5 cysts (13.2%) were in active stage (CE1: 4 and CE2: 1), 5 cysts (13.2%) were transitional (CE3A: 2 and CE3B: 3), and 28 (73.7%) were inactive (CE4: 8 and CE5: 20). **DISCUSSION/CONCLUSIONS:** A second US screening will be carried out in the Ñorquinco or Comallo areas (Río Negro, Argentina), to individuate the highest and lowest prevalence villages/areas where sampling of environmental matrices will be used carried out. This work is supported by EU-LAC Health project and National funding agencies of the participating institutions - PERITAS project (<http://eulachealth.eu/>).

**Keywords:** International public health, ultrasound screenings, pathways of transmission

### ULTRASOUND SCREENINGS IN LIMARÍ PROVINCE, CHILE: PRELIMINARY RESULTS FROM STAGE 1 OF PERITAS PROJECT.

Gerardo ACOSTA-JAMETT\*, Natalia CASTRO, Francesca TAMAROZZI, Leonardo UCHIUMI, Juan Carlos SALVITTI, Edmundo LARRIEU, Saul SANTIVANEZ, Maria J PERTEGUER, Mar SILES-LUCAS & Adriano CASULLI

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**INTRODUCTION:** PERITAS project is aiming to elucidate the pathways of transmission of cystic echinococcosis (CE) by two sequential stages to be carried out in Chile, Argentina, and Peru. **STAGE 1** is a cross-sectional ultrasound (US)-based prevalence study to identify endemic villages/areas where the following **STAGE 2** case-control study will be implemented at household and community level, detecting and comparing matrices contamination with eggs/DNA of *Echinococcus granulosus sensu lato*. **MATERIALS AND METHODS:** During the first stage of this project we surveyed by abdominal US people from nine areas with varying degree of urbanization residing in the Monte Patria municipality, Limarí province of Coquimbo region, Chile. In total, we examined 2,435 inhabitants older than 5 years of age. **RESULTS:** Overall, 39 (1.6%) cases were CE positive (38 with cysts in the liver and 1 in the kidney). Average age of cases was 55.5 years, and one cyst was found in a 10-years-old kid. Prevalence ranged from 0.5% to 2.7% in seven areas and in two high prevalence areas values were 5.2% (12/230) and 6.7% (8/120). 15 cases (39%) were in people currently inhabiting urban sites. Prevalence in urban areas was 1.0% (15/1480) and 2.5% (24/955) in rural sites. 16 cysts (41%) were in active stage (CE1:16) and 18 were inactive (CE4:11 and CE5:7), 4 were transitional (CE3a and CE3b:2 each stage); finally, 1 person has 3 cyst in different stages: CE3a/CE4/CE5. **DISCUSSION/CONCLUSIONS:** This is the first abdominal US survey for CE carried out in Chile. We have successfully detected areas with high and low prevalence where environmental sampling will be conducted to assess differences in risk of exposure to environmental contamination with *E. granulosus* in the next phases of this project. This work is supported by EU-LAC Health project and FONIS T020067- PERITAS project (<http://eulachealth.eu/>).

Ultrasound screenings in Communities of Junin, Peru: Preliminary results from stage 1 of PERITAS project  
Saul Santivañez (Peru)

Abstract is not available.

## Symposium 9 WHO-IWGE

Reinserting CE and AE in the New NTD roadmap and Universal Health Coverage  
Bernadette Abela-Ridder PhD (WHO, Geneva, Switzerland)

Abstract is not available.

The 'CE Technical Manual': reaching out to health services and patients  
Thomas Junghans MD (Germany)

Abstract is not available.

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Update from WHO-IWGE - AE Clinical group. Building the AE Technical Manual  
SurLaurence Millon PhD (France)

Abstract is not available.

Surveillance and Control of Cystic Echinococcosis  
Edmundo Larrieu PhD (Argentina)

Abstract is not available.

IWGE subgroup Control of CE  
Marshall Lightowlers (Australia)

Abstract is not available.

AE control, an update  
Paul Torgerson PhD (Switzerland)

Abstract is not available.

International consensus on terminology in the field of echinococcosis  
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Introduction: The lack of terminology standardization in the numerous disciplines related to echinococcosis has long been a source of misunderstanding between scientists, and between scientists and professionals, At the 27<sup>th</sup> World Congress on Echinococcosis in 2017, a multidisciplinary working panel was launched in order to reach a consensus on echinococcosis terminology in English. Materials and Methods: According to the 'Formal Consensus' methodology, 3 groups, comprising representatives from 15 countries, were constituted: a Steering and Writing Group (SWG), a Consultation and Rating Group (CRG), and a Reading and Review Group (RRG). Their remit was to work on 3 main areas: 'Species and epidemiology', 'Biology and immunology', and 'Clinical aspects'. Consultation of the CRG on a list of 137 terms established by the SWG generated a literature review and free discussion between the participants, and led to the constitution of 3 lists: a) an

agreement for approval; b) an agreement for rejection; and c) debated issues submitted to a poll. Participants in the CRG then had to rate each word and expression, and the consensus reached was based on the median of the votes, and complementary discussion where necessary. Definitive tables of the approved terms were included in a paper published by the journal PARASITE after review by the RRG. Results: Following these extensive deliberations, 9 species of *Echinococcus*, currently described on genetic grounds, and the names of the 3 main human diseases caused by *Echinococcus* spp. – ‘cystic echinococcosis’ (CE), alveolar echinococcosis (AE) and neotropical echinococcosis (NE) – were definitively approved. It was agreed that the adjective ‘hydatid’ should be reserved for cases of *E. granulosus sensu lato* only. A number of confusing and misleading terms were rejected and a list of the recommended terms is now available online. Conclusion: This consensus terminology will serve as the basis for regular updates and future work by groups of specialists in the echinococcosis field, and for instigating similar recommendations in the various languages of the endemic countries.

## Symposium 10 EcoHealth / OneHealth

One Health, The Reality out There. Translating theory into practice

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The One Health approach is crucial to managing and protecting the health of humans, livestock, wildlife and the environment. However, its ultimate success comes only when the proposed recommendations are embraced and implemented not only by government officers but all the stakeholders, including local communities. While we continue to claim that we understand the One Health concept and how to implement it, it is often only applied at the central governmental level and in academic settings or research teams using external funding. Over the years, various multidisciplinary teams including policy makers researchers and academicians helped look at the One Health approach in a holistic manner and design ways for its implementation; yet the One Health approach is currently still undermined by:

- 1) Funding deficiencies, especially at the inter-sectoral level, and substantial differences in funding between human and animal health programs.
- 2) Silo-ization in the missions of governmental and intergovernmental institutions and agencies. Significant gaps remain in the implementation of One Health at the local level where diseases are prevalent.
- 3) Disparities in the education, training, and job opportunities at the human-animal interface.
- 4) Sustainability concerns and competing priorities.

By solving these challenges, we can make the One Health approach a more powerful tool for prevention and control of zoonotic diseases. In this paper, the authors discuss

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challenges of the One Health approach and outcomes using lessons learned from their own experience in countries where they have worked.

Echinococcosis in Europe: transmission ecology, diagnosis and control in a Swiss “One Health” network

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Neglected helminthic zoonoses such as alveolar (AE) and cystic (CE) echinococcosis are persisting worldwide and even re-emerging in several European countries. There are various reasons that may account for these trends – such as socioeconomic changes, the close emotional tie between people and companion animals, and ecological changes. Fox populations have increased in many regions of Europe, and high population densities are observed today also in the middle of many conurbations across Europe. At the same time, recreational environments are increasingly designed closer to natural ecological systems. Such areas provide good shelter for foxes and boost populations of voles that represent urban reservoirs for zoonotic parasites, including *Echinococcus multilocularis*. Understanding parasite transmission and the risks of human infection requires regionally well adapted transdisciplinary investigations involving wild animal ecologists, medical and veterinary experts, as well as epidemiologists. An important outcome of such research programmes was the finding of a highly scattered transmission risk for AE. Transmission hotspots were identified in recreational urban peripheries where foxes intensively predate on susceptible intermediate hosts and cause a high environmental contamination with *E. multilocularis* eggs. Significant research has accumulated in the last decades and has laid the basis for the development of transmission blocking control strategies, which have been validated for rural and urban situations. However, economic and health considerations of the health authorities of Switzerland resulted in the political decision for not taking direct action against AE. Uniform recommendations for the control of parasites of dogs have been developed for Europe on a non-governmental level ([www.ESCCAP.org](http://www.ESCCAP.org)) and serve as continuous education of veterinarians as well as instructions for pet owners.

One Health and the evolving epidemiology of *Echinococcus granulosus* in Australia.

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There have been two major events that have led to the reduction of infection with *Echinococcus granulosus* in Australian domestic dogs, especially those living in rural areas. The patents for the highly efficient cestocidal drug, praziquantel, expired in the 1990s and following expiry, praziquantel was included in a range of cheap deworming products for dogs, available widely in Australian supermarkets and rural produce stores. The second important event was the development of highly palatable, nutritionally-balanced dry dog food that is convenient to store and feed to dogs. As a consequence of these developments, the prevalence of hydatid disease in sheep has fallen markedly. However, *E. granulosus* has not gone away, the parasite has become firmly embedded in our wildlife, transmitting mainly between wild dogs (dingoes and dingo/domestic dog hybrids) and macropod marsupials. This wildlife reservoir is

perpetuating the existence of the parasite in Australia and acting as a widespread, ongoing source of infection to domestic livestock, domestic dogs and potentially also humans. More recently, *E. granulosus*-infected wild dogs have begun encroaching into urban centres. This change in wild dog behaviour is exposing people in some urban communities, hitherto considered safe from infection, to possible exposure with *E. granulosus*. The 'One Health' implications of these changes in the epidemiology of *E. granulosus* in Australia will be discussed.

## SCIENTIFIC ORAL PRESENTATIONS

Geospatial disease modelling to identify potential environmental determinants of human echinococcosis in Kyrgyzstan.

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Introduction: Human echinococcosis is a neglected zoonosis that has become a public health emergency in recent years in Kyrgyzstan. In this country, we previously identified significant risk hot spots of the two main forms of the disease—cystic echinococcosis (CE) and alveolar echinococcosis (AE). Given that CE and AE transmission risk to humans can be linked to environmental and climatic determinants, in this study we aim at further exploring these associations in a spatial modelling framework. This framework is meant to enable assessing risk factors explaining the spatial distribution of CE and AE, and, ultimately, predicting locations where control interventions could be efficiently employed. Materials and methods: We computed average annual standardized incidence ratios (SIRs) of CE and AE at the local community level in Kyrgyzstan between 2014 and 2016 using data from the Kyrgyz national surveillance system. To account for the long latency of both disease forms, we obtained environmental and climatic raster data for the years 2000, 2005, and 2010. We then averaged the cell values of each raster at the local community level, and tested the resulting variables for correlations with the SIRs of CE and AE using the Spearman's rank correlation test. Variables exhibiting significant correlation were then used to fit a Bayesian conditional autoregressive spatial model of SIRs of CE and AE. Results: Our preliminary results will be presented at the conference. Discussion: Our modelling framework has the advantage of including selected explanatory variables as well as spatially structured random effects. This enables to

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include information on spatial processes (e.g. spatial autocorrelation) that have not been captured by the selected explanatory variables by improving the overall fitting process. Conclusion: Our results provide insights into environmental and climatic determinants that could play a role in Echinococcus transmission in Kyrgyzstan. Further research on the disease ecology is needed to support human echinococcosis control.

Key words: Human echinococcosis, spatial analysis, environment

A One-Health evaluation of the socio-economic impact of cystic echinococcosis in the Veneto region, a hypoendemic area in Northern Italy

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Introduction: Cystic Echinococcosis (CE), a worldwide public health hazard, has been emerging in the Veneto region (Italy), with confirmed circulation in animals and records of human cases. The objective of this research project was to evaluate the economic impact of CE in this region using a One Health perspective. Materials and Methods: An integrated epidemiologic and economic model, and a data collection strategy were developed based on literature review and expert consultation. The main epidemiologic and economic outcomes of the disease on human, animal and environmental health were identified. The national and local health authorities were consulted and provided data on human cases and on treatment costs. The number of sheep and bovines found infected at slaughterhouse were obtained through data mining at concerned private and public institutions. Costs due to offal condemnation and decreased productivity were estimated according to literature. Results: 234 hospitalized human CE cases of residents of the region (144 foreigners and 90 Italians) were recorded in the period 2005-2016, corresponding to an average of 19.5 cases/year, without increasing or decreasing trend in the period. The majority (n=129) were treated surgically, whereas 105 patients were hospitalized for medical treatment. In Veneto, the cost of the clinical management ranged from €2,600.00 to €10,160.00, depending on cyst localisation (liver or lungs) and type of treatment (surgical or medical). It was not possible to identify with certainty the autochthonous nature of the infections in humans. In animals farmed in the region (an average of 4,786 sheep and 59,355 dairy cattle slaughtered yearly), the estimated prevalence of CE was 6% in sheep and 0.2% in dairy cattle. Discussion and Conclusion: The impact of the disease in animals was found to be negligible. The few human cases imply a relatively important cost for the regional health system, despite the uncertainty on the origin of the infection. An effective surveillance system in animals is desirable, to limit the risk of increasing autochthonous human cases.

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# Symposium 11

## Diagnosis of Echinococcosis in definitive host

Update on knowledge of transmission of Echinococcus to humans

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Humans are dead-end hosts for Echinococcus spp acquiring the infections by uptake of viable parasite eggs from contaminated/unwashed food sources; or via exposure by hand-mouth contact to eggs derived from the environment (coat of definitive hosts). In fact, Echinococcus spp has been recently claimed to be an important food-borne parasite, however, this still needs confirmation as, for example, is not possible to know if all eggs found in food sources are viable. Contamination rates of taeniid eggs between 0.9 and 18.3% have been reported in vegetables from different parts of the world. However, to date, there is no standardized methodology for detecting taeniid eggs in food samples. Even more, the relative importance of the different mechanisms of transmission remains unknown because they are difficult to quantify and because they vary between and within endemic areas. For example, in some endemic areas, there is no availability/consumption of fresh produce; therefore, the most likely way of transmission can be the hand to mouth after close contact with an infected dog. Molecular detection/identification of Echinococcus infection in different matrices has improved enormously in recent years. However, the detection of Echinococcus in soil specimens is much less efficient than with faeces. Recent studies have detected DNA of Echinococcus spp in food, soil and also in water. One main concern of such studies is the lack of inclusion of test to investigate if any egg found in these matrices is actually viable. To date, there is no in vitro test for assessing the viability of the eggs of Echinococcus species. This presentation aims to highlight key points in which there is a lack of knowledge on the transmission of Echinococcus to humans, data regarding a standardized method for detection of Echinococcus and other taeniids in food sources for human consumption is also presented.

Key words: Echinococcus, transmission, foodborne

Surveillance of CE in DOGS  
Edmundo Larrieu (Argentina)

Abstract is not available.

Cystic Echinococcosis: environmental diagnosis  
Graciela Santillán

Cystic Echinococcosis is a parasitic zoonosis, which is caused by environmental biological contamination with *Echinococcus granulosus* eggs. In rural habitats the most polluted sites are those where dogs spend the longest time of the day, for this reason it is considered that the epidemiological unit, is the microenvironment of the rural housing. In it dogs become infected and in turn contaminate it. Hence, the strategy that would best be adapted to control and monitor the parasite would be the activities in the areas

surrounding of the homes of endemic areas. In these areas, the domestic cycle is established by contact of the definitive host with the parasitic viscera of the intermediate slaughtered hosts, that's to say, zoonotic cycle and between the definitive host and the environment, that's to say environmental cycle, being the man an intermediary host of the environmental cycle. The eggs and remains of the parasite after being released with feces, are left in the environment at the mercy of the factors that mobilize and move them actually producing biotope contamination. In the Department of Parasitologia of INEI, a coproELISA was developed for the detection of parasite-specific antigens in the feces. This technique uses polyclonal antibodies against adult tapeworm antigens, parasite antigens can be detected after eleven days p.i, before rupture of the gravid proglotid. It has the disadvantages that can cross-react with other parasites. For this reason, molecular diagnostic techniques were developed to confirm the positive results. CoproPCR consists of amplifying a partial sequence of the mitochondrial gene CO1. This tool allows you to differentiate eggs from different tapeworms. Both methods have the advantage of being able to process a large number of samples with little risk to the operator and the environment. The PCR technique is also used for the detection in soil, plants and water of parasitarian material. The positive reaction in samples taken from the environment surrounding the home suggests the extent of environmental pollution in a dwelling located in the endemic area.

#### SCIENTIFIC ORAL PRESENTATION

Development and validation of a Copro-ELISA sandwich for detection of *Echinococcus granulosus*-soluble membrane antigens in dogs

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Cystic echinococcosis (CE) is a parasitic zoonosis caused by the larval stage of the tapeworm *Echinococcus granulosus*. Detection of the adult stage in the canine definitive host is essential for estimating infection rates, surveillance and monitoring of CE control programs. This study sought to develop and validate a coproantigen sandwich enzyme linked immunosorbent assay (Copro-ELISA), based on antibodies against *E. granulosus* soluble membrane antigens (EGMA), that is capable of distinguishing infected and non-infected dogs. Anti-*E. granulosus* polyclonal immunoglobulin G antibodies were obtained from rabbit antiserum against EGMA. Pre-adsorption step with faeces of non-infected dogs was not included. Optimization of the test was performed with 51 positive and 56 negative stool samples of canine echinococcosis. Specificity, sensitivity, cross-reactivity, intra- and inter-assay precision, and over time detection were evaluated. According to the receiver operating characteristic analysis, the diagnostic sensitivity and specificity were 96.1% (CI: 85.9–99.6) and 98.2% (CI: 89.5–100), respectively. Negative and positive predictive values were 96.5% (CI: 91.7–100) and 98% (CI: 94.1–100), respectively. No cross-reactivity with *Taenia hydatigena*, *Dipylidium caninum* or *Toxocara canis* was observed. Intra- and inter-assay repeatability showed values of less than 15% of the variation coefficient. The over time detection in dogs was from 20–27 days post-infection with *E. granulosus*. The Copro-ELISA based on EGMA detection offers a simplified in-house development of diagnostic testing. This assay showed high specificity and sensitivity and had no cross-reactivity with other parasites. Further studies and development of this test in a kit format may be useful for the detection of active infection in dogs living in CE endemic regions.

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## Symposium 12

### Neotropical Echinococcosis: emerging zoonosis

Epidemiological situation and importance of the Neotropical Echinococcosis in America and the world

<sup>1,2</sup>Katherina A. Vizcaychipi, <sup>3</sup>Natalia Casas, <sup>4</sup>Nilton Giotti Siqueira, <sup>5</sup>Rosangela Rodrigues, <sup>6</sup>Michael Lise Laurence, <sup>7</sup>Karen DeMatteo, <sup>8</sup>Oscar Noya Alarcon, <sup>9</sup>Guzman Sienna, <sup>10</sup>Cesar Gavidea, <sup>11</sup>Sofía Duque Beltran, <sup>1</sup>Ariel Naidich, <sup>2</sup>Esteban Couto, <sup>12</sup>Larissa Zanette, <sup>12</sup>Marco Vigilato, <sup>13</sup>Leonardo Uchiumi, <sup>14</sup>Miguel Rinas.

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Neotropical echinococcosis (NE), polycystic, caused by *Echinococcus vogeli* and unicystic, caused by *Echinococcus oligarthra*, is considered an emerging zoonotic disease in 15 countries of Central and South America (Nicaragua, Costa Rica, Panamá, Colombia, Venezuela, Suriname, Guayana Francesa, Ecuador, Brasil, Perú, Bolivia, Chile, Paraguay, Uruguay, Argentina) due to its advances in morbidity and mortality characteristics, as well as its eco-epidemiological dimension. *E. vogeli*, with an environmental niche determined by the distribution of its host (species - specific), is the most important in Public Health. Within the framework of the Sustainable Development Objectives and with the One Health approaches this presentation aims, to disseminate knowledge of the most important aspects of their situation, the advances generated in terms of strategies for prevention, surveillance and control in the countries involved, and above all, its aims to raise awareness from the motivation to work with integrated approaches, which allow to optimize the information channels and lead to become aware of the real dimension of the NE.

Treatment of polycystic hydatidosis  
Katherina Vizcaychipi, MSc (Argentina)

Abstract is not available.

NEOTROPICAL AND CYSTIC ECHINOCOCCOSIS, A MULTIDISCIPLINARY VIEW FROM THE PUBLIC HEALTH PERSPECTIVE

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Neotropical echinococcosis (NE) differs from cystic echinococcosis produced by *E. granulosus* (CE) in terms of geographical distribution, epidemiology and life cycles. NE is maintained in a predation/selvatory cycle between *Speothos venaticus* and *Cuniculus paca* (*E. vogeli*) and between felids and rodents (*Dasyprocta*) (*E. oligarthrus*). Also, as is the case with CE, domestic dogs are primarily responsible for human infections with *E. vogeli*. Human populations that coexist with wild canines and felids are more exposed to the risk of acquiring NE. The objective is to disseminate and know the epidemiology of both echinococcosis and the importance of surveillance, control and integration of regional control programmes. The echinococcosis can be reported to the National Health Surveillance System. Argentina has a CE Surveillance and Control Program, so it is essential to take advantage of efforts and resources, integrating all echinococcosis. When carrying out surveillance, prevention and control activities in the field, the NE should also be taken into account. The main activities would be: a) ultrasound and serological cadastres of the human population, b) collection of canine and *S. venaticus* fecal samples and wild cats for environmental surveillance, c) notification and analysis of organs of dead carnivores and rodents, d) distribution of veterinary antiparasitic drugs, e) educational activities, f) control of animals at border crossing points, g) training of health teams, tourism personnel and park rangers. There should be intersectoral work among all the Ministries (Health, Agriculture, Environment, Tourism and Education), with multidisciplinary work being important, giving priority to primary health care. An integral approach to echinococcosis is fundamental for the control of this zoonosis in a country.

## SCIENTIFIC ORAL PRESENTATION

Neotropical polychistic echinococcosis in *Cuniculus paca* (Linnaeus, 1766) and *Dasypus novemcinctus* (Linnaeus, 1758) in the high basin of the Itaya River, Peru

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*Echinococcus vogeli* originates neotropical polycystic echinococcosis (NPE) (CIE-10,XXII: U51X), an emerging zoonotic disease in tropical countries of Central and South America. Its natural cycle is adapted to a mandatory relationship between *Speothos venaticus* and one of its main prey, the majaz (*Cuniculus paca*), which is, in turn, one of the most consumed species as a source of animal protein in the region. The aim of this work is to know the frequency of *E. vogeli* in intermediate hosts of communities of the upper basin of the Itaya River. Descriptive design, period 2016–2017, carried out in the communities of Melitón Carbajal, Luz del Oriente, 28 de Enero and Nueva Villa Belén. The research was authorized by the Executive Directorate of Forestry and Wildlife. The methodology for

collecting samples was based on subsistence hunting carried out regularly by local communities. A total of 8 volunteer hunters were identified, 2 hunters in each community. The collaborating hunters were trained in the post-mortem recognition and storage of cystic/polycystic masses found in thoracic-abdominal organs of shot animals. These actions were integrated into a participatory wildlife management program. Macroscopic, morphometric and molecular analysis was performed. Polycystic masses were presented in liver of collected from majaces (11.1%; 4/36) and from an armadillo (*Dasypus novemcinctus*). Based on the morphological classification of species, the size, shape and proportions of the protoscolex rostellar hooks obtained from the majaces and the armadillo are compatible with *E. vogeli*. The molecular analysis of one majaz confirmed *E. vogeli*. This work contributes to the eco-epidemiological knowledge of and allows confirming the first reported of *E. vogeli* in armadillo in the Peruvian Amazon and second report in South America, helping to reinforce the emergent character of the NPE.

Keywords: Neotropical Polycystic Echinococcosis – *Echinococcus vogeli* – *Cuniculus paca* – *Dasypus novemcinctus* – Itaya River, Peru

## SCIENTIFIC SESSION 3 (oral presentations)

Control of Hydatid Disease in the Context of the Neglected Zoonotic Disease System: Applying One Health in the Oloisukut Conservancy, Narok, Kenya

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Cystic echinococcosis control has generally been isolated from that of other diseases and inter-sectoral collaboration has always been a challenge. Cystic echinococcosis is only one of the neglected zoonotic diseases (NZDs) that have been prioritized by the WHO and by the Kenyan Government. Livestock keeping communities are constantly exposed to several human and animal diseases that require integrated control measures in order to save resources. The objective of this project is to facilitate collaboration between the main stakeholders and use innovative approaches such as the One Health, the horizontal and participatory approaches in order to:

1. Identify the main health issues of people and animals and the risk factors that enable their perpetuation in the Oloisukut Conservancy.
2. Design and implement adapted control measures to address these health issues in humans, animals and the environment in a holistic manner, ensuring collaboration of the main health professionals and using cost effective measures.
3. Evaluate the impact of these interventions in the community.

Surveys, focus groups and interviews have been conducted to assess the general needs of the population and to collect data on the diseases. Samples are being collected from people, animals and their environment and analyzed to estimate the disease prevalence. The results will be used as baseline data for control. Initial investigations showed that NZDs and other human/animal diseases are highly prevalent. The lack of water, sanitation, and health and education structures contribute greatly to the perpetuation of these health issues and need to be addressed. An integrated control program would be a great benefit to the community. The one health and the horizontal approaches to disease control should be considered, as recommended by the WHO, in communities that

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are challenged by the scarcity of water, health and education structures and government funding. Preliminary results will be presented.

Eg95 vaccine monitoring of intermediate hosts. Analysis and inclusion of the socio-cultural context in vaccination programs.

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Introduction: Hydatidic disease is linked to symbolic and social considerations and responds to a cultural logic that interacts with a set of traditional knowledge and practices that need to be understood. Vaccination of intermediate hosts contributes to reduce the level of transmission of *E. granulosus* and thus, reduce the incidence of human infections, even when control programs address many practical difficulties. In order to validate a model supporting the One Health approach, that might be reproducible successfully in different regions of South America, we raise the socio-cultural understanding and the analysis of the environment and the operational conditions, looking for a mediation according to the context and using a special vaccination methodology in each level of difficulty. Materials and Methods: In controlled trials immunization of sheep (0, 30 days and annual booster) with EG95 vaccine available in South America were carried out. After experimental challenge with *E. granulosus* eggs, subsequent necropsy, and the percentage of reduction of the viable cysts versus EG95 antibodies level were calculated. Control Programs included: Chubut 2007-2013-Río Negro 2009-2015- Alto Bio-Bio, Chile 2016-2020.

Key Words: EG95 Vaccine, control programs, social- cultural context

Epidemiological factors associated with human cystic echinococcosis investigated through a semi-structured questionnaire during the ultrasound surveys of the heracles project

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Patrizio PEZZOTTI, Loredana G POPA, Valeri VELEV, Mar SILES-LUCAS, Enrico BRUNETTI, Adriano CASULLI F TAMAROZZI: WHO Collaborating Centre for the epidemiology, detection and control of cystic and alveolar echinococcosis (in animals and humans), Department of Infectious Diseases, Istituto Superiore di Sanità, Rome, Italy.

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INTRODUCTION: Risk factors for human cystic echinococcosis (CE) are poorly defined. They have been explored through questionnaires in hospital-based and field-based studies, mostly testing samples of limited sizes and being highly heterogeneous in terms of case definitions and data collected, providing contrasting results. We present the analysis of risk factors questionnaires administered to participants during the large HERACLES study (2014-2015), which identified abdominal CE by ultrasound, applying a stringent case definition. MATERIALS and METHODS: A semi-structured questionnaire was administered to 24,687 people from rural Bulgaria, Romania, and Turkey. Questions

concerned demography, CE awareness, occurrence of cases in the family, dog and livestock-related practices, and food- and drinking water-related habits. Variables associated with CE infection at  $p < 0.20$  in bivariate analysis were included into a multivariable logistic model, with a random effect to account for clustering at village level. Adjusted odds ratio (AOR) with 95%CI were used to describe the strength of associations. Data were weighted to reflect the relative distribution of the rural population in the study area by country, age group and sex. RESULTS: Valid records from 22,027 people were analysed. According to the main occupation in the past 20 years, "housewife" (AOR 3.11 [1.51-6.41]) and "retired" (AOR 2.88 [1.09-7.65]) showed a significantly higher risk of being infected compared to non-agricultural workers. "Having relatives with CE" (AOR 4.18 [1.77-9.88]) was associated with higher odds of CE infection. Dog-related and food/water-related factors were not associated with infection. DISCUSSION/CONCLUSIONS: Our results point to infection being acquired in a "domestic" rural environment and support the view that CE should be considered more a "soil-transmitted" than a "food-borne" infection. Community-specific and habits-specific questionnaires, and studies on parasite contamination of matrices, are needed to shed light on actual sources of infection and at-risk behaviours. This work was supported by the EU FP7, HERACLES project (GA602051).

Keywords: cystic echinococcosis, potential risk factors, questionnaire

#### Identification of potential 'hot spots' of cystic echinococcosis transmission in the province of Río Negro, Argentina

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Introduction: cystic echinococcosis (CE) is a parasitic zoonosis produced by endemic *Echinococcus granulosus* (EG) in the province of Río Negro, Argentina, where social, cultural, and environmental and livestock production conditions support a favorable epidemiology to perpetuate the cycle of transmission. Surveillance actions within the control program (based on the deworming of dogs with praziquantel), focus since 2003 on annual ultrasound screenings in school children from 6 to 14 years (identification of transmission in the recent past) and five-year cross-sectional studies for determine fields (epidemiological units or EU) with parasitized dogs (identification of current transmission) by coproELISA with confirmation by Western Blot or PCR.

Objective: to identify risk areas for human populations, through the application of analytical models with added evidence from all surveillance sources to concentrate control activities.

Materials and methods: the information was spatially added in 13 program areas, each with its head hospital, and, on a smaller geographical scale, in 80 Primary Health Care Centers (CAPS) that provide coverage to 100% of the dispersed population in an area of 1200013km<sup>2</sup> of Patagonian steppe. CoproELISA: included 3 randomized samplings (2003/5, 2009/10, 2017/18) in 780 UE, 1790 samples of canine fecal matter were obtained, while 34515 screened with ultrasound were grouped accumulated for the periods 2003/08, 2009 / 16 and 2017/18. Heat maps were constructed at the smallest geographic scale with QGIS 3.4.6.

Results: The prevalence for the three samplings with coproELISA were 14.7%, 12.1% and 7.8% of EU with infected dogs, while with ultrasound the prevalence of infected schoolchildren was 0.4%, 0.2% and 0.1%, respectively, delimiting the areas with greater transmission.

Discussion: there is little information on the temporal-spatial relationship between sources of human-animal surveillance in EQ. The developed models show the temporal-spatial distribution at a geographic scale that allows to adjust the control strategies to the areas of influence of the CAPS where the possibility of transmission to man is maintained within a framework of sustained decrease in prevalence.

### Human Echinococcosis, Kyrgyzstan, 2001–2018

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Human cystic and alveolar echinococcosis. Transmission to humans occurs when eggs of the tapeworm, excreted by the final hosts (usually dogs, foxes), are accidentally ingested. The larva's primary target organ is the liver, lung where it proliferates slowly, but the larva also spreads into extrahepatic structures and even metastasizes to distant organs. Methods: Case Retrieval Case detection and data collection have been organized according to the existing infrastructure of the national health system. Patient data are sent to the national registry, where they are controlled and approved for electronic recording. 2. Case Definition Diagnosis of alveolar echinococcosis is confirmed by positive histopathology and typical liver lesion morphology identified by imaging techniques (ultrasound scan, computed tomography, and magnetic resonance imaging). Positive serologic results without suggestive imaging findings or positive histopathology do not qualify for a case definition. 3. Case Report Form and Completeness of Registration Questionnaires are used which have the epidemiological part, which the patient must answer, and the clinical part, which the attending physician must fill out. In addition to demographic baseline data, information was collected related conditions: a place of residence and occupation. Results: In the last 18 years in Kyrgyzstan, there has been a strong increase in the incidence of echinococcosis. For 2018 the incidence of CE was 15.1/100,000 and AE 2.0 / 100,000. The real incidence of the disease may be considerably higher as many people may not have access to appropriate medical services. Of particular concern is the steady increase in the incidence rate amount of children and young people, reaching 14-18 % of the number of patients admitted for surgical treatment. Conclusion: Conclusion. In connection with the urgency of the problem of echinococcosis, it is necessary to study and develop an optimal system of epidemiological surveillance of echinococcosis on the basis of comprehensive studies.

### CYSTIC ECHINOCOCCOSIS IN CATTLE OF MISIONES, ARGENTINA

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In Argentina, the intermediary host that has the greatest epidemiological importance of cystic echinococcosis (CE) is the sheep but bovine livestock production it is importance in the economy, being able to substantially influence the incidence of this disease. The province of Misiones, since three decades ago shows an increase in the cattle activity. The aim of this work is to know the cases of bovine echinococcosis found in slaughter in the province of Misiones. The data presented for livestock evolution and animals slaughtered with lesions compatible with echinococcosis for the period 2003-2018 were obtained from the National Service of Agri-Food Health and Quality (SENASA). Information of macroscopic lesions compatible with hydatid cysts was also obtained from a private abattoir of Misiones. Parasitological confirmation was performed by microscopic and molecular analysis. Regarding livestock evolution, in 2003 were registered 24.884 heads and in 2018 there is an increase of 443.491 bovine heads. SENASA reports an average of 10.2% of bovines with compatible lesions with echinococcosis. From the private abattoir, were analyzed the viscera of 150 cattles belonging to eight farms, from the departments of Apóstoles, Capital, Ruiz de Montoya and Leandro N. Alem. The 6,6% corresponded to hydatid cyst (3 cows, 1 cow, 6 steers). Of the cysts obtained, 2 were fertile, 5 infertile, 2 calcified and 1 abscessed. The location by affected organ was: pulmonary, renal, hepatic; with diameters from 1 to 3,5 cm. The measurements of the rostellar hooks were morphologically compatible with *E. granulosus*. The molecular analysis confirmed *E. granulosus s.s.* G1 and *E. ortleppi* G5. The results of the study illustrate the increase in cattle activity in the economy of this region confirming the occurrence of CE in the cattle. This work reaffirms the importance of epidemiological and parasitological studies of the disease, allowing the strengthening of the provincial and national surveillance system.

Keywords: Echinococcosis - cattle - Misiones, Argentina.

#### Social Representations on Echinococcosis cystic of families of High Mountain, Tucumán, Argentina.

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Introduction: The Cystic echinococcosis (EQ), is a parasitic disease caused by *Echinococcus granulosus*. Definitive host is the dog and complete its life cycle in other animals. It represents a serious public health problem in livestock areas. Tucuman endemic region corresponds to high mountain, with a population of 1067, families scattered and without basic services. Between 1997-2016 had a prevalence of 565 inhabitants cases/year/100.000. To control the (EQ) is important the health education, directed to amend customs, beliefs, and attitudes from the affected communities, contributing to its transmission and maintenance; and they can be linked to social representations on (EQ) or perception subjective personal and/or family of those who get sick. Know them, is the main objective of the present work. Methodology: qualitative, retrospective study with interviews open to families and individuals affected by (EQ). Analysis using the grounded theory (Glaser and Strauss) Results: the emerging

categories were: 1) Exclusion: caused by lack of protection before the inaccessibility to health services, with a perception of neglect in terms of the role of the State. (2) Change on a daily basis: reflected in familiar destructure, uprootedness, separation of the land and the affects, loss of belongings, caused by the internment in strange areas (city). (3) Fear of death: the cultural vision leads to the personification of disease and terror of the unknown, derived from invasive procedures and bloody. (4) Health policies that ignore them: the population generally unknown rights in health; and it has no accessibility practices of greater omplexity. Discussion/Conclusion: there is no bibliography allowing you to compare the results obtained in the study. This uncommon approach about the subjectivity of the patient against the disease, highlights the fears, helplessness, the feeling of abandonment and fear of death, of those affected by this neglected disease.

Keywords: echinococcosis, perception, social representations

#### Economic impact of hydatid disease on the Australian beef industry

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Hydatid disease in the Australian beef industry is believed to have a major financial impact due to the condemnation and downgrading of infected offal. The aims of this study were to determine the prevalence and economic impact of hydatid disease in an eastern Australian beef abattoir. Meat inspection data from 1.1 million cattle slaughtered between January 2011 and December 2017 were collected from an eastern Australian abattoir. Summary statistics were used to describe the organs infected with hydatid disease and prevalence as reported by routine meat inspection. The direct losses resulting from the condemnation and downgrading of offal infected with hydatid cysts were estimated overall and using data stratified by age, sex and feed-type. Prices received for organs were those obtained by the abattoir and applied as uniform distributions. Monte Carlo simulation techniques were used to estimate the financial impact to the abattoir. Reported prevalence of hydatid disease in the study population was 8.9% (95% confidence interval [CI] 0.89–0.90). Most infected cattle had cysts in the liver, lungs or both (17%, 4%, and 77%, respectively). The estimated total losses incurred by the abattoir for the study period due to condemnation and downgrading of infected offal were AU\$681,021 (95% CI AU\$564,806–813,944). The median annual loss was AU\$97,289 (95% CI AU\$80,687–116,278). The estimated financial loss at an individual animal level was AU\$6.96 per infected animal (95% CI AU\$5.77–8.32). The estimated losses to the abattoir in this study were considerable, indicating that losses to the Australian beef industry would likely also be large. Given the results, further studies should be conducted to estimate the financial losses in other Australian abattoirs to determine industry economic impact, and to investigate the cost-benefit of control programs for hydatid disease in Australian beef cattle.

#### LUNG HYDATIC CYST OF CHILDREN

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**Introduction:** Hydatidosis is a common disease in Algeria. In children, it's the first localization. The positive diagnosis is based on epidemiological, clinical, biological and radiological arguments. Surgical treatment with its various techniques remains the most effective. **Method:** The purpose of our study is to analyze the radio clinical aspects and evaluate the different therapeutic modalities of pulmonary hydatidosis in children who have been treated in the thoracic surgery department during a period of three years from June 2015 until December 2018. We recorded 15 cases ranging in age from 5 to 16 years old. Most of the patients came from rural areas. **Results:** The techniques used are puncture-aspiration and enucleation technique of Ugon. No radical surgery was noted in our study. The immediate postoperative course was good and long-term radiological and biological monitoring is still on going to detect recurrence or other localization. **Conclusion:** hydatid disease is a benign pathology but its occurrence in a child explains the high rate of parasitic infestation which always reminds us that prevention remains the most effective therapeutic weapon.

### The specificity of the surgical management of the lung hydatid cyst

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**Introduction:** Hydatid cyst disease is a parasitic infestation that is endemic in many parts of sheep and cattle farming and it is still a real public health problem in Mediterranean countries. The lung is the second most frequent localization. It's usually asymptomatic and discovered incidentally. Currently, the treatment of lung hydatid cysts LHC involves surgery (conventional surgery or video surgery), the percutaneous drainage (PAIR) of cysts has not proved effective, given the high complication rate. We'll discuss the main surgical techniques practiced on the lung in our department, especially the cuneiform resection which allow to obtains a very good results. **Methods:** we reviewed the records of 102 patients operated for pulmonary hydatid cyst in our department during 5 year period (2014-2018). **Results:** The LHC essentially affected young people. All these patients benefited the cuneiform resection. In our series the morbidity was 15% and the mortality was low (1%). **Conclusion:** The hydatid cyst surgery is always based on principles of bases that are: complete removal of the cyst (if feasible), protection of the operative field, prevention of bronchus damage and the management of the residual cavity. And our technique gave us very good results especially in children.

**Key words:** Hydatid cyst, Children, Surgery, Residual cavity, Cuneiform resection

## Scientific Session 4 (oral presentations)

Distribution of alveolar echinococcosis according to environmental and geographical factors in Germany, 1992-2018

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**Introduction:** Alveolar echinococcosis (AE) is a rare zoonotic disease caused by the larval stage of *Echinococcus multilocularis*. In the present cohort study, we analyze the distribution of alveolar echinococcosis according to environmental and geographical factors in Germany. **Material and Methods:** We identified the place of residence of 591 cases of AE from the national database for alveolar echinococcosis, and georeferenced these localities in the Universal Transverse Mercator coordinate system. Data on elevation, temperature, precipitation and land cover were mapped out and correlated with the distribution of cases of disease during the period 1992-2018. Differences in distribution between the groups in the elevation, temperature and precipitation models were analysed with the Kruskal-Wallis test. Moran's I statistic was used for spatial autocorrelation. p-values < 0.05 ( $\alpha = 0.05$ ) were considered statistically significant. **Results:** A total of 591 (100.0%) cases of AE were included in the study, consisting of 259 (43.8%) men and 332 (56.2%) women. The overall prevalence of AE was 0.71/100,000 inhabitants. The mean age of the patients was  $63.63 \pm 18.67$  years. Based on the case definition of AE 289 (48.9%) of the cases were 'confirmed', 155 (26.2%) 'probable' and 147 (24.9%) were 'possible'. The results showed a heterogeneous distribution of AE cases with a higher concentration in southern Germany ( $I = 0.225517$ ,  $Z = 35.8182$  and  $p < 0.001$ ). There was a statistically significant difference between the geographical and environmental factors (precipitation, temperature, altitude, land cover) and the distribution of cases in Germany ( $p < 0.0001$ ). In regions with higher altitudes (505-672 m (m a.s.l.)), moderate average temperatures (13-14 °C) and higher precipitation rates (5-7 mm) most cases of disease were recorded. **Discussion / Conclusion:** In Germany there are statistically significant differences in frequency distributions of AE according to environmental and geographical factors.

**Keywords:** Alveolar Echinococcosis; *Echinococcus multilocularis*; Environmental and geographical factors

#### Quantitative Study of Fat Deposition and Iron Deposition in Patients with HAE by HISTO and q-DIXON Techniques on MRI

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**Objective:** to analyzed the liver iron deposition and fat deposition in hepatic alveolar echinococcosis (HAE) patients with HISTO and q-DIXON imaging, and correlation analysis with serum ferritin. **Methods:** 15 cases with HAE and 20 healthy volunteers were enrolled in this study. Q-Dixon and Histo sequence were performed on 3.0T MR scanner. The HISTO, could automatically scan the selected region of interest the area under the curve which means the fat fraction was calculated and draw the curve, and R2

value of the target also can be displayed. The measurement of the region in Dixon imaging of interest should be close to ROI of HISTO. Then, measured the fat fraction and the R2\* value in fat fraction maps and R2\* relaxation maps. Results: The R2\* value of HAE group and normal group were (133.05±99.74) and (88.74±52.77), The R2 value of HAE group and normal group were (39.98±8.71) and (51.07±15.94). Most patients in the HAE group had higher R2\* value and R2 value than the normal group. Statistically significant differences in R2\* value and R2 value were found between two groups. A positive correlation was found between R2\* value and serum ferritin (P<0.05) in HAE group. There is no statistically significant differences in R2\* value and R2 value were found in HAE group. A positive correlation was found between R2 value and serum ferritin in HAE group. The HISTO and Dixon sequences measured liver fat changes in patients without fat deposition. Conclusions: The iron deposition in the liver of patients with HAE is higher than healthy volunteers, and their fat deposition was basically normal. HISTO and Dixon imaging were sensitive to iron deposition and fat deposition, they can be used as an effective method for non-invasive and quantitative evaluation of HAE iron deposition.

#### Health-related quality of life in patients with alveolar echinococcosis: surgical versus conservative drug therapy – a quality of life comparison

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Alveolar echinococcosis (AE) is a rare zoonosis caused by the parasite *Echinococcus multilocularis*. Aim of the study was to compare HRQoL in patients with AE depending on their therapeutic regimen namely conservative drug treatment with long-term benzimidazoles versus surgical therapy by resection of the parasitic liver lesions. Material and Methods:

The 36-Item Short Form Health Survey (SF-36) questionnaire, including other echinococcosis-related questions, was used to measure HRQoL. The SF-36 scales were evaluated according to the algorithms provided by the authors. The statistical analysis was performed with SAS version 9.2. The significance level was set at  $\alpha=0.05$ ,  $p<0.05$  corresponds to statistical significance. Results: The investigated group consisted of conservatively drug treated (n=30) and patients with curative surgery (n=25) with confirmed AE. The study was performed at an infectious disease outpatient department from April 2018 to October 2018. The conservatively drug treated patient group consisted of 15 men (50.0%) and 15 women (50.0%) with an average age of 55.7±16.7 years (Median: 59). The surgery group consisted of nine men (36.0%) and 16 women (64.3%) with an average age of 53.3±31.9 years (Median: 54). The physical quality of life of the conservatively drug treated patients did not show any significant differences to the surgical treated group (45.2 ±11.4 vs. 47.6 ±9.9;  $p=0.4079$ ). There was also no significant difference regarding the mental quality of life between the conservatively drug treated

patients, and those treated with curative surgery ( $45.5 \pm 10.6$  vs.  $47.3 \pm 10.8$ ; 0.5206). Nevertheless, there was a slight advantage in the physical and mental scores of the patients treated with surgery. Discussion / Conclusion: The evaluation showed no statistically significant differences in HRQoL in patients with AE dependent on the applied treatment strategy (conservative drug versus curative surgical therapy).

Keywords: Alveolar Echinococcosis; health-related quality of life; Therapy

Clinical Research of Shear Wave Elastography (SWE) in Differential Diagnosis of  
Hepatic Alveolar Echinococcosis (HAE) and Hepatocellular Carcinoma (HCC)

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**Objective:** To evaluate the sonographic features of real-time shear wave elastography (SWE) in patients with hepatic alveolar echinococcosis (HAE) and hepatocellular carcinoma (HCC) and investigate the clinical application of SWE in differential diagnosis of HAE and HCC. **Methods:** There were 20 cases in the HAE group and 30 cases in the HCC group in the study. All the cases were performed SWE before surgery, the mean elasticity of Young's modulus of all the cases were obtained. All the lesions were conformed by pathologies as the gold standard. SWE results were compared with the pathologies to evaluate the accuracy and efficacy of using SWE to diagnose the HAE and HCC. **Results:** The mean elasticity of Young's modulus of the lesions was ( $34.57 \pm 10.76$ ) Kpa in the HAE group and ( $21.82 \pm 3.51$ ) Kpa in the HCC group. The mean elasticity of Young's modulus of lesions had significant difference between HAE and HCC groups ( $P < 0.001$ ). The critical point of the mean elasticity of Young's modulus of the lesions between HAE and HCC was 27.43 Kpa by ROC curve, the sensitivity was 80%, and the specificity was 97%. The area under the ROC curve was 86.9%. **Conclusion:** SWE has an important value of clinical application in diagnosis between HAE and HCC.

**Keywords:** shear-wave elastography ; Hepatic alveolar echinococcosis ; Hepatocellular Carcinoma ; ROC curve

*Echinococcus multilocularis* infection in solid-organ transplant patient recipients

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**Introduction:** Alveolar echinococcosis (AE), a severe helminth infection, was reported to have a higher incidence, a faster course and a more difficult diagnosis in immunosuppressed hosts. We aimed to study cases of de novo AE in solid-organ transplant (SOT) recipients. **Methods:** We looked for de novo AE cases in SOT recipients in the Swiss Transplant Cohort Study and in the FrancEchino registry for the 01/2008-08/2018 period. We collected data on the clinical presentation, treatment and course of AE. **Results:** Eight patients were identified, 6 in France and 2 in Switzerland (kidney=5, lung=2, heart=1). AE presented as abdominal pain in 4 cases and was otherwise asymptomatic at diagnosis. The location of AE was the liver in 7 cases and the lung in one case. The median interval between transplantation and diagnosis was 66 months (range 12-230). For two patients, CT and US revealed no lesion 43 and 26 months prior to diagnosis, respectively. Em2+ was positive in 1 out of 3 cases, Em2 in 1 out of 2, EgHF in 4 out of 5 and IHA in 3 out of 4. Western Blot was positive in all cases. MRI was performed in 7 patients, with atypical results in 2 cases. Five patients were operated on. One complete resection was achieved. Two patients died following operation. A non-operated patient died 5 months after diagnosis. Albendazole was given to all patients and was well tolerated. Median follow-up duration was 15 months (range 0-49). AE remained stable in 3 cases, regressed in 1 and progressed in 1. **Conclusion:** AE in SOT recipients was associated with high mortality (37.5 %). The diagnosis was often incidental and made difficult because of negative serologies. Imaging was sometimes atypical and Western Blot could confirm the diagnosis. SOT recipients should have a specific follow-up in endemic areas.

#### Human Alveolar Echinococcosis in the Czech Republic: Clinical Outcome and Laboratory Parameters

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**Introduction:** Human alveolar echinococcosis (AE) is an emerging zoonotic infection in Central and Eastern Europe. The objective of our study is to describe clinical and laboratory characteristics, management and outcome of patients with AE treated in our departments. **Method:** This is a prospective descriptive study of fifteen AE cases treated at the Departments of Infectious Diseases in Hospital Na Bulovce in Prague and Regional Hospital Liberec since 2012. Epidemiological, clinical and laboratory parameters has been collected and evaluated in all patients. **Results:** There have been managed total 15 patients (10 women, 5 men) with AE at our departments since 2012. All cases were acquired in the Czech Republic (14) or Slovakia (1). The age of patients was between 7 and 73 years at the time of diagnosis. Lesions were localized in liver only (11 patients), in liver with propagation to adjacent organs (3) and in liver with multiple metastasis in lung

(1). One patient acquired infection after liver transplantation. Serology for echinococcosis was positive (11) or borderline (3) in all patients, except one infected after liver transplantation. There was not significant eosinophilia, but total IgE was highly elevated in 6 of 15 patients. Detailed results of hematology and immunology parameters will be presented. The treatment is based on long-term continuous albendazole administration, which was not tolerated in one patient, that is treated with mebendazole. In addition, 8 patients underwent liver resection and 2 patients liver transplantation. Clinical status of all patients is favourable, except for one who died from AE unrelated reason. Conclusions: AE has increasing incidence in the Czech Republic with 3 to 5 new cases diagnosed each year. Diagnosis of AE is difficult as infection can be asymptomatic for many years. Management of AE must be multidisciplinary, radical surgery is followed by long-term albendazole administration.

#### Long-term cryopreservation of *Echinococcus multilocularis*

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**Introduction:** *Echinococcus multilocularis* isolates are maintained at laboratories typically in rodents by serial passages of the metacestode material (interval of 2-3 months). Cryopreservation presents a better alternative to replace serial passaging for strain maintenance purposes. To determine whether cryopreservation of *E. multilocularis* metacestode stage would be a viable option for long-term maintenance, different isolates were cryopreserved in 1984-86. The aim of the current study was to test the viability of *E. multilocularis* metacestode material after being cryopreserved for 35 years in liquid nitrogen. **Material and methods:** Isolates (from Switzerland Alaska, St. Lawrence, Canada) cryopreserved either as small tissue fragments (STF) or tissue blocks (TB) with either 5% dimethylsulfoxide (DMSO) or 10% glycerol as cryoprotectant, were thawed in a 37°C water bath, washed in culture medium and cultivated in vitro. Protoscolex movements and proliferation of vesicles were recorded. Furthermore, 39 gerbils were infected through an intraperitoneal injection to test for the potential of metacestode proliferation. Additionally, haplotypes were determined for the isolates based on *cox1*, *cob* and *nad2* genes. **Results:** Samples from Switzerland corresponded to the European, from St. Lawrence to the North-American, and from Canada to the Asian haplotype cluster. Protoscolexes showed movement in vitro in 87.5% of STF samples, and in 67% of TB. Comparing the two different cryoprotectant substances, movement was recorded in 75% of samples frozen with DMSO, and in 78% frozen with glycerol. Vesicles were present in vitro in altogether 6 out of 17 samples, of these 5 were in samples preserved as STF. In vivo experiment is currently ongoing. **Conclusion:** Preliminary results of the current study indicate that the larval stage of *E. multilocularis* is able to survive long-term cryopreservation (at least 35 years), and the cryopreserved material can be used to resume the cycle in vitro and potentially in vivo for experimental use. **Keywords:** *Echinococcus multilocularis*, alveolar echinococcosis, cryopreservation

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Worldwide literature on epidemiology of human alveolar echinococcosis: a systematic review of research published in the twenty-first century.

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**Introduction:** Human alveolar echinococcosis (AE) is a potentially lethal zoonosis caused by the cestode *Echinococcus multilocularis*. The aim of this systematic review is to establish a comprehensive global AE literature overview taking into account the epidemiologically relevant AE research of the twenty-first century. **Material and Method:** We systematically searched the global literature published from 2001 through 2018 via MEDLINE, EMBASE, the Russian databases eLIBRARY.RU, CyberLeninka, the Chinese databases CNKI, VIP, Journals. ac.ir (Farsi language-based), Jordan E-Library (Arab language-based) and supplementary Google Scholar, in accordance with the PRISMA guidelines. QGIS software was used for the mapping of the affected countries. **Results:** We have listed 154 relevant publications in the final literature synopsis in consideration of our quality assessment. Including non-autochthonous cases, human AE was reported in 36 countries within the northern hemisphere from 2001 to 2018. The first publication of AE in Tajikistan, Pakistan, South Korea, Belgium, the Netherlands, Slovakia, Hungary, Lithuania, Latvia, Slovenia and Morocco occurred in this century; further first cases in Taiwan, Thailand, and Denmark were considered to be non-autochthonous by the authors. The highest total case numbers ( $n \geq 100$  in a single article) were reported in France, Germany, Switzerland, Poland, and Lithuania, including China and Kyrgyzstan with by far the highest prevalence figures. **Discussion/Conclusion:**

Our paper emphasises the increasing spread of reported cases and the rise in its numbers in the literature of the twenty-first century, especially in western, northern and eastern Europe, as well as in central Asia. Epidemiological studies on human infections are lacking in many parts of the world.

**Keywords:** Alveolar echinococcosis, *Echinococcus multilocularis*, distribution, epidemiology, prevalence, maps

**29 DE OCTUBRE****PILOT FIELD TRIAL OF THE EG95 VACCINE AGAINST OVINE CYSTIC ECHINOCOCCOSIS IN RIO NEGRO, ARGENTINA: LAST INFORMATION**

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Introduction: Cystic echinococcosis (CE) is endemic in the Rio Negro province of Argentina. The objective of the study was to assess the inclusion of the EG95 for sheep in the control program. Materials and method: An intervention study was defined comprising, in total, an area of 5820 Km<sup>2</sup>. Lambs received two vaccinations with the EG95 vaccine followed by a single booster injection when the animals were 1-1.5 years of age. Vaccination of lambs born into one trial site was introduced and continued until today. Evidence for *Echinococcus granulosus* transmission was monitored before and after vaccination by coproELISA in faecal samples of dog, necropsy on adult sheep and by ultrasound screening in schoolchildren. Results: 31894 doses of vaccine were applied between 2009 and 2018, which a vaccination coverage of 80.1%/85.7% (57.3% average for fully vaccinated). Before the introduction of the vaccine 56.3% of the 6-year-old sheep were infected at necropsy and 84.2% of the farms had infected sheep; and with coproELISA 9.6% of dog fecal samples were positive and 20.3% of the farms had infected dog. After the vaccine was introduced, 21.6% of sheep older than 6 years were found to be infected at necropsy and 20.2% of the farms were found to be infected; in dogs, with coproELISA 3.7% of samples were positive, with 8.9% of farms having a positive dog. In 2016 was diagnoses the last case of *E. granulosus* infection diagnosed by US screening in a 6-14 years old child with 0 new symptomatic case in 2016/9. Discussion: Included in the analysis are discussions of difficulties experienced in the field which affected correct vaccine administration as well as social features and practices that may impact on echinococcosis control and the EG95 vaccination program in Rio Negro.. Vaccination was effective even in a difficult, remote environment where only approximately half the lambs born into the communities were fully vaccinated.

**MOLECULAR-EPIDEMIOLOGICAL STUDIES ON PATHWAYS OF TRANSMISSION AND LONG LASTING CAPACITY BUILDING TO PREVENT CYSTIC ECHINOCOCCOSIS INFECTION: OUTLINE OF PERITAS PROJECT**

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**INTRODUCTION:** The WHO advocates the control of echinococcosis and general hygiene education is envisaged among control strategies for cystic echinococcosis (CE). Although ingestion of contaminated food/water and direct contact with infected dogs are classically mentioned as infection sources, the precise sources and at-risk behaviours for human infection have not been clearly individuated. Addressing specific at-risk behaviours is expected to increase the effectiveness of hygiene education. PERITAS project aims to elucidate through an integrated, systematic approach the pathways of transmission of *Echinococcus granulosus*. **MATERIALS and METHODS:** The study design is a cross-sectional prevalence study of human infection followed by a case-control molecular epidemiology study on several matrices, comparing level of contamination in high vs low endemic areas. Prevalence of human CE in active stages will be identified by population-based ultrasound surveys in selected areas of Argentina, Chile, and Peru. Sampling of different matrices in these identified areas will be performed, followed by application of molecular techniques to identify contamination and species/genotype of the parasite and an ad-hoc questionnaire focused on specific habits and behaviours will be provided to participants. In addition, biological samples will be collected to feed the biological repository Echino-Biobank, and training and feed of the International Register of CE (IRCE), will allow long-lasting capacity building. **RESULTS:** The analysis of the data obtained in clusters of active human CE infection will allow the identification of matrices contaminated by *E. granulosus* eggs and of at-risk behaviours/habits associated with odds of CE infection. **DISCUSSION/CONCLUSIONS:** The direct applicability of project results will allow the implementation of more precisely-targeted human-centred control interventions, saving time and costs of control activities and increasing the efficacy of control programs for the prevention of new human infections. This work is supported by EU-LAC Health project and National funding agencies of the participating institutions - PERITAS project (<http://eulachealth.eu/>).

**Keywords:** cystic echinococcosis, pathways of transmission, environmental contamination.

#### THE PREVALENCE OF LIVER ECHINOCOCCOSIS OF PUPILS IN BANMA DARI, QINGHAI

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**Objective:**The aim of our study was to observe the prevalence of liver echinococcosis in banma and dari areas of Qinghai. **Methods:** B-mode ultrasound was applied to scan the livers and the patients with suspected hydatid mass would be drawn venous blood to check the antibody of echinococcosis. The reference standard was used with "Diagnostic criteria of liver echinococcosis (WS257-2006)". **Results:**956 pastoral areas of Tibetan students ageing from 3 to 17 years were investigated. 19 students were finally diagnosed suffering from hepatic echinococcosis. Three students were newly found. The overall prevalence rate was about 1.99%. 16 cases were suffering from alveolar echinococcosis and the prevalence rate was 1.67%. Three cases were suffering from cystic echinococcosis and the prevalence rate was 0.32%,Alveolar echinococcosis prevalence rate was significantly higher than that of cystic echinococcosis prevalence, the difference was statistically significant (  $\chi^2 = 8.98$ ,  $P = 0.002$  ) . **Conclusion:** The prevalence rate of

liver hydatid disease in Dari and Baima areas was still high, mainly with hepatic alveolar echinococcosis. Early detection and early treatment is vital. Health propaganda and education and vigorously promote knowledge of liver hydatid disease prevention should be widely carried out in these areas. The control of the source of infection and dogs and stray dogs worms expelling and epidemic prevention treatment are also the focus of hydatid disease prevention and control.

Key Word: Liver echinococcosis Epidemic Ultrasound

**COLOSTRAL IMMUNITY INDUCED BY EG95 RECOMBINANT VACCINE IN SHEEP  
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**Introduction:** The Eg95 recombinant vaccine protects sheep against cystic echinococcosis. Ruminants have a syndesmochorial placenta and transplacental transfer of macromolecules is not allowed. In neonates, serum immunoglobulins (Ig) levels are no detectable at birth. Transfer of Ig from blood into mammary secretions, starts several weeks prepartum and maternal antibodies are transferred to lamb by passive immunity exclusively through colostrum. **Objective:** Correlation between Eg95 antibody levels (Eg95-Ac) transferred to colostrum and the age of newborns whose mothers had received EG95 vaccine during pregnancy. **Materials and Methods:** Two groups, M1 (n=4) and M2 (n=5) of Merino sheep reared at the Research Center for Zoonosis received 1 and 2 doses of commercial Eg95 vaccine respectively. Serum and colostrum/milk samples were collected after birth and during 28 days in each group. Serum samples from C1 (n=5) and C2 (n=6) groups, belonging to newborn lambs from M1 and M2 respectively, were obtained immediately after birth and during 90 days. Colostrum/milk and serum specimens were tested for EG95-Ac by enzyme-immunoassay. **Results:** M2 and C2 groups induced enhanced titers of specific Eg95-Ac, significantly higher than those obtained in M1 and C1. The maximum level of Eg95-Ac was detected in colostrum from M2 immediately after delivery. In C1 and C2 newborns, Ac-EG95 levels was not detected before the first ingestion of colostrum, reaching a peak during the first 24 hours after birth. Increased and sustained titers of Eg95-Ac were detected in the serum samples from C1 and C2 lambs for more than 4 weeks. **Discussion:** Our results confirm that Eg95-Ac level increase significantly in colostrum prior to delivery, achieving an effective transfer of specific antibodies to newborns. Vaccination of lambs should be started while maternal Eg95-Ac are detected in blood, a recommended immunization schedule would include a first dose of Eg95 vaccine near the first month of life, a second dose 30 days later, and an annual booster.

**Keywords:** Cystic echinococcosis, Eg95 vaccine, colostrum immunity

**THE EVALUATION OF TEN-YEAR LABORATORY RESULTS OF CYST HYDATID PRE-DIAGNOSED PATIENT SAMPLES ANALYZED IN MICROBIOLOGY LABORATORY OF ESOGÜ MEDICAL FACULTY HOSPITAL BY AGE, GENDER AND RELEVANT DEPARTMENTS.**

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**Objective:** Cystic echinococcosis (CE) is zoonotic metacestode infection caused by Echinococcus granulosus whose adults are identified as hydatid worm and effects herbivorous animals and humans. Diagnosis of CE is based on clinical symptoms, imaging techniques and detection of specific antibodies, especially in patient sera. The

aim of this study was to evaluate the relationship between ten years serologic laboratory records of patients with pre-diagnosis of CE according to age, sex and referral departments. Materials and Methods: Total of 892 patients who were sent to Eskisehir Osmangazi University Faculty of Medicine Microbiology Laboratory with the pre-diagnosis of CE from various clinics in the last decade were studied with *E. granulosus* IgG ELISA test according to the recommendations of the company. In the evaluation of the results, cut-off and above values were accepted as positive and the experiment was repeated for the limit values and suspicious positives. The results were analyzed with pearson chi-square. Results: Of the 892 CE pre-diagnosed patient's samples, 482 ones have belonged to females (54%) and 410 have belonged to males (46%) Seropositivity has been found in the ratio of 116 of females and 89 of males (p 0,669). The average of age was divided into two groups with cut-off ratio (35,41) and seropositivity was found to be higher under the age of 35 (p <0,000). According to logistic regression analysis, the risk of developing the disease decreases by approximately 0.01 times with the increase by one unit in age. When the units with a percentage less than 0.5 were excluded for the distribution of the results by their clinics, the p value of the dependency test was found to be p <0,000. Seropositivity was mostly defined in the Polyclinic of Radiology with 68.2% and in the Polyclinic of Chest Diseases in accordance with the distribution of organ involvement (p <0,000). All results are indicated in the tables. Conclusion: Although the incidence of CE depends on age and outpatient variables, the risk was found to be higher in younger ages. No gender differences were observed. Although there are many studies on animals on CE in our country, there is little data on prevalence in humans. The disease is an important zoonosis that continues to affect human health in our province and its surroundings as in our country. The retrospective study presented here is designed to raise public health awareness.

Key Words: Echinococcus granulosus, Retrospective, Hydatid Cyst

#### URBAN CANINE ECHINOCOCCOSIS SURVEY IN THE PROVINCE OF RÍO NEGRO, PATAGONIA, ARGENTINA

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In Río Negro province, Cystic echinococcosis is an endemic zoonosis produced by *Echinococcus granulosus*, which has sanitary importance in rural communities. Recently, presence of this parasite has been recognized in dogs from Bariloche city. The objective of this work was to evaluate canine echinococcosis in three urban areas in the south of the Province. Samples of canine feces were taken in Bariloche (207), Valcheta (53) and Sierra Colorada (12). These cities have an estimated population of 120000, 4000 and 2000 inhabitants, respectively. A sandwich ELISA test with polyclonal antibodies was used. All samples were processed in duplicate, values of optical density (DO) higher than 0.235 for both trials were considered positive. Additionally, in Bariloche and Valcheta, surveys to dog's owners were conducted to determine the habits of dogs and responsible pet ownership. Positive canine feces for *Echinococcus* sp were found in Bariloche (4.8%), Valcheta (20.7%) and Sierra Colorada (16.7%). More than 50% of dogs roams freely in all

cities, with a mean density of dogs/house ranging from 2.2 (Bariloche) to 4.2 (Valcheta). Most dogs had been dewormed more than 10 months ago. In Valcheta and Sierra Colorada, half of the respondents performed slaughter at home or in their farm. Dogs from Valcheta and Sierra Colorada have a closer relationship with rural areas, since they are near to sheep farms, which would explain the higher infection values. Although the province has a dog deworming program in rural areas, surveys showed that deworming is insufficient in the cities. These results would indicate that cities constitute infection foci which, in general, are not taken into account. Educational programs of responsible dog ownership are needed to control urban transmission of Cystic echinococcosis.

Key words: Canine echinococcosis, urban areas, Patagonia

#### FASCIOLA HEPATICA CO-INFECTION IS ASSOCIATED WITH MORPHOLOGICAL CHANGES IN ECHINOCOCCAL CYST OF NATURALLY INFECTED CATTLE

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**Introduction:** Polyparasitism, when animals harbor more than one parasite concomitantly, is a common and understudied event in animals worldwide. *Fasciola hepatica* and *Echinococcus granulosus* are both common helminths of ungulates and can be frequently found co-infesting cattle, however the implications of this polyparasitism not well documented. The metacestode of *Echinococcus granulosus*, is composed of a cellular layer called germinal layer, an extracellular matrix called laminated layer surrounded by the host immune response called adventitial layer. The adventitial layer in cattle is composed of a granulomatous immune reaction and is involved in the Echinococcal Cyst (EC) fertility, that is, the ability to produce protoscoleces and complete the life cycle. Due to the systemic immune modulating abilities of *Fasciola hepatica*, we hypothesize that co-infection generates a synergic relationship between both parasites. **Materials and Methods:** We sampled cattle lung and liver ECs from adult animals in abattoirs and performed morphohistological analysis in slides stained with haematoxylin-eosin. **Results:** As expected, co-infected animals display more frequently an absence of lymphoid follicles and palisading macrophages, both hallmarks of the granulomatous immune response, while also having thicker and organized laminated layers. Conversely, co-infection is associated with lower intensities of infection and smaller ECs, especially in lung infertile cysts and thinner laminated layers in liver. Although we found no fertile cysts in the liver of co-infected animals, co-infection is not associated with EC fertility. **Conclusion:** We conclude that *Fasciola hepatica* co-infection with *Echinococcus granulosus* has a paradoxical effect, as it is both synergistic and antagonistic in naturally infected animals. **Funding:** FONDECYT N° 1190817.

Keywords: polyparasitism, hydatid cyst, *Fasciola hepatica*.

#### FOLLOW-UP OSSEOUS CYSTIC ECHINOCOCCOSIS IN URUGUAY

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**Introduction:** Cystic echinococcosis (CE) is a neglected chronic disease in humans, second in ranking in the worldwide list of foodborne parasites, and representing one of the 17 neglected tropical diseases prioritized by the World Health Organization. Characteristically, CE lesions are found in the liver and the lungs, but virtually any part of the body might be affected i.e, spleen, kidneys, heart, central nervous system and bones among others. However, the incidence of bone involvement in CE is currently estimated to be between 0.5% and 4%. **Methodology:** A longitudinal study between 1990 and December 2018 was conducted on patients presenting with osseous CE and treated with pharmacological/chirurgical protocols at the National Institute of Orthopedics and Traumatology; the Medical Treatment of Echinococcosis Reference Unit, and the Department of Parasitology and Mycology at the School of Medicine, UdelaR, Montevideo, Uruguay. Epidemiological, diagnostic, clinical, and therapeutic data was collected from patients with a long-term follow-up. **Results and Discussion:** During the study period, 18 of the 91 patients with CE exhibited pathological bone involvement (20%). The most frequently affected areas were the spine, followed by iliac and pelvic, coccyx, femur and tibia. The organs most commonly affected were the liver and lungs. The most prevalent presenting symptom was intense pain followed by medullar syndrome, pathological fractures. All of the studied patients underwent, at least once, surgery for osseous CE removal. Subsequently, all received albendazole monotherapy or in combination with praziquantel. **Conclusions:** The diagnosis and management of CE with bone involvement has been lately reevaluated with most cases now considered as a chronic phase of the disease. Pre-surgery pharmacological treatment allows, in the vast majority of cases, a significant decrease in cyst size, good debridement and laminectomy. Our data suggests the necessity of, case by case, lifelong continuous follow-up to prevent relapses and potential sequelae.

**ERANET-LAC PROJECT: DEVELOPMENT OF NEW DIAGNOSTIC AND TREATMENT OPTIONS FOR HELMINTHIC NEGLECTED DISEASES (NDTND)**

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**INTRODUCTION:** Our project aims at developing new therapeutic and diagnostic tools to contribute to the control of neglected diseases caused by helminthic parasites, such as cystic (CE) and alveolar (AE) echinococcosis. **MATERIALS AND METHODS:** Specific *Echinococcus* molecules are being evaluated as new therapeutic/diagnosis targets using bioinformatics, molecular biology, biochemistry and biophysical methods integrated with relevant clinical and epidemiological information. **RESULTS:** The evaluation as therapeutic targets of highly expressed, specific molecules that may exert essential functions for the parasite, such as microRNAs and lipid binding proteins is underway. The small RNAs and proteins actively secreted by larval stages of these parasites are being characterized to assess their potential as biomarkers. Ultrasound surveys and sera collection from humans affected by CE are being performed in order to link the potentially new biomarkers in the stage specific approach according to WHO-IWGE (Informal Working Group on Echinococcosis). **CONCLUSION:** The expected outcomes of the project

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are the development of new compounds that bind and inhibit essential and unique molecules of these parasites, and to find new detection tools to improve the status of both treatment and early diagnosis of these complex and neglected parasitic diseases. This work was supported by ERANet-LAC 2<sup>nd</sup> Joint Call (<http://www.eranet-lac.eu>) and the National funding agencies of the participating institutions - NDTND project.

KEY WORDS: cystic and alveolar echinococcosis, microRNA, Lipid Binding Proteins.

### CYSTIC ECHINOCOCCOSIS IN BOLIVIA, PRESENT AND UNKNOWN

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**Introduction:** The knowledge of the current situation of cystic echinococcosis (CE), caused by *Echinococcus granulosus sensu lato*, in Bolivia, is restricted to a handful of publications in English, which have highlighted that CE is present and causing an unknown number of cases. However, to date is not clear what the current situation of CE in the country is. **Material and method:** We reviewed all documents, written in Spanish and English, including scientific publications in local journals, government reports and thesis investigating CE in Bolivia. **Results:** CE has been diagnosed and reported in Bolivia since the first half of the XX century. Ever since data from a number of case reports and some retrospective studies have suggested that CE human cases have actually increased since the '70s and '80s in Bolivia. In total, 789 CE human cases have been reported in the literature until 2018. Furthermore, data from the '70s reported up to 50% of dogs infected in some communities near La Paz. On the other hand, data informing prevalence in livestock animals remains a mystery due to the lack of the systematic gathering of official reports for the confiscation of infected viscera in official abattoirs. **Discussion/conclusion:** The numbers of cases reported in Bolivia suggest that there is enough evidence to acknowledge that CE is a problem in Bolivia which needs further investigation to understand the real impact of the disease in the population. Such baseline data can inform policymakers in the country to establish activities which can prevent the persistence of the transmission of the parasite and decrease the number of human CE cases in the future.

Key words: Bolivia, cystic echinococcosis, review

### ANALYSIS OF PEDIATRIC HYDATID CYST PATIENTS IN ESKIŞEHİR OSMANGAZI UNIVERSITY HOSPITAL -TURKEY, DURING 2012- 2019

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**Background:** Cystic echinococcosis (CE) is an important health problem in childhood period. We aimed of this study was to evaluate clinical and laboratory characteristics of CE in children in Eskisehir/Turkey. **Materials and Methods:** CE data of 51 children aged 1-18 years who were admitted to the pediatric surgery clinic of ESOGU Faculty of Medicine from 2012 to the present day were examined in terms of various variables; clinical findings, diagnostic methods, age, gender, location and number of the cyst, surgical method, and risk factors were reviewed. **Results:** The age range of the cases was between 1-18 and the most common age was 11-15 years (49%). The distribution by gender

was found to be 30 (59%) boys and 21 (41%) girls. Hepatic involvement in 51%, lung involvement in 33%, both lung and liver involvement in 10%, and other organ involvement in 6% were reported. All of the cases were supported by at least one radiological diagnosis. Echinococcus granulosus IgG ELISA test used in laboratory diagnosis was studied in only 35 patients. Seropositivity was detected in 22 cases (63%) and seronegativity in 13 cases (37%). Cough, abdominal and chest pain, fever, weakness are the most common symptoms. The most common treatment method was cystectomy and excision and PAIR was performed in 4 patients. Preoperative and postoperative drug follow-up was performed in 41 patients. There was no difference between risk factors and CE transmission risk. Conclusion: CE is a serious public health problem in childhood in developing countries. In recent years, there has been an increase in childhood CE cases in around our city. Seroepidemiological studies are important in endemic areas. In addition to supporting suspicious clinical findings and radiological examinations, it should be supported by rapid reliable serological tests in the follow-up of the operated cases.

**Key words:** Cystic echinococcosis, Childhood, Diagnosis, Turkey

#### DIAGNOSTIC PERFORMANCE OF MACROSCOPIC EXAMINATION FOR DETECTION OF HYDATID CYSTS IN SLAUGHTERED SHEEP

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Diagnosis of Cystic Echinococcosis (CE) in sheep relies essentially on necropsy findings. Clinical symptoms, if present, may be easily overlooked while the use of immunological tests is still not recommended for intra vitam diagnosis. The present study aimed to assess the diagnostic performance of macroscopic examination for post mortem diagnosis of ovine Cystic Echinococcosis (CE). In situations where there is no single, accurate reference standard a Bayesian statistical approach can combine the results of multiple analytical tests in order to obtain diagnostic performance. To this purpose livers and lungs were sampled from 102 sheep after slaughter and examined by three methods: macroscopic examination, histopathology and a PCR assay. Macroscopic examination consisted of direct observation, palpation and incision of the organs. The presence of hydatid fluid, germinal layer and protoscolices were also evidenced to confirm the diagnosis of CE. Hydatid cysts were excised and divided for both histopathological examination and molecular analysis. Histology sections were stained with Hematoxylin-Eosin and PAS to evidence the presence of an outer PAS-positive acellular laminated layer with an inner cellular nucleated germinal layer, considered to be a suggestive pattern of metacestodes of the Echinococcus. A PCR was performed for amplification of the calreticulin gene for detection of *E. granulosus sensu stricto*. Data from the three diagnostic methods were analyzed to evaluate sensitivity (SE), specificity (SP), positive predictive value (PPV) and negative predictive value (NPV) of macroscopic examination for CE diagnosis using a Latent Class Analysis model. Macroscopic examination showed a SE of 98.3 (95% CI, 93.3-99.8), a SP of 98.8 (95% CI, 95.4-99.5), a PPV of 97.7 (95% CI, 78.4-100), a NPV of 99.5 (95% CI, 96.6-100). Our results confirmed the excellent diagnostic performance of the method suggesting that the experimental design used in this work can be used for validation protocol in quality assurance system.

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## COEXISTING PULMONARY HYDATID DISEASE AND TUBERCULOSIS

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**Introduction:** Algeria is an endemic country for hydatid disease and pulmonary tuberculosis. The association of this two pathologies is not exceptional. We will show you the cases of 11 patients who had been operated for hydatid cyst without knowing that they had underlying tuberculosis. **Methods:** More than half of the patients were children. 9 patients benefited from conservative surgery and there were 2 cases that had lobectomy pulmonary. **Results:** The postoperative course was long with more than 10 days of hospitalization and above average morbidity (23). It is the pathological anatomy of the post operative specimen which revealed the tuberculous disease underlying the hydatid disease. **Conclusion:** In endemic areas, we must think about the combination of these two pathologies, to better manage these patients postoperatively. **Keys words:** Hydatid cyst, tuberculosis, surgery

## ECHINOCOCCOSIS IN AÑATUYA, SANTIAGO DEL ESTERO, ARGENTINA: INTENSIVE DIAGNOSTIC WORK-UP

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Echinococcosis is a zoonosis related to the contact with dogs in areas of animal production. Diagnosis is usually based on epidemiological data together with imaging results and serology. The objective of the present study was to perform early detection in a community from Añatuya, Santiago del Estero. The epidemiological situation in this community is favorable for the transmission of the disease. School N° 28 "Hernando de Magallanes" from the rural community of Miel de Palo was used as the base for the performance of the US study and the collection of blood samples. This study was approved by the Ethical Committee from the Ministry of Health of Santiago del Estero. A total of 183 people participated in the study by giving their consent for the performance of the US and collection of blood. Serum samples were analyzed through an indirect enzyme-linked immunosorbent assay as a screening and subsequent confirmation through Western Blot was performed. Through the ELISA and WB, nine samples were found to have specific antibodies. The US showed a cyst compatible with Echinococcus in the liver, one with vesicular lithiasis and another one with hepatomegaly (these participants were treated with albendazole). Serological tests are complementary to the US and serve to detect exposure to the parasite since not all individuals that are exposed to Echinococcus sp. eggs get infected or the infection is aborted before the production of cysts, this is related to host factors. Another reason for positive serology is the presence of cysts in other sites not covered by the US. In those cases (one was not performed due to pregnancy), X rays were performed. The X rays showed a lack of lung cysts. The results obtained suggest that strategies for the prevention and control of Echinococcosis are necessary in this area.

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DETECTION OF ECHINOCOCCOSIS IN COMMUNITIES OF THE CALCHAQUÍ VALLEYS.  
SALTA ARGENTINA

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In the Calchaquíes valleys, goats breeding are carried out by small producers. The basis of the economy is agriculture, production of artisanal cheeses and sale of kids and adult goats. According to the account of the owners have been observed cysts in the liver and lungs of the goats slaughtered, observing dogs in all the flocks. The aim of the work was to make a diagnosis of the situation in the municipality of Payogasta, where it is suspected the circulation of *Echinococcus granulosus sensu lato*. This municipality is located 14 km from Cachi and 151 km from Salta. The flocks are bred between the 2300 and 2900 meters above the sea level. A transversal survey, it was made between June and October of 2018, in 20 goat family units (GFU). The producers are owners of small fields no more than 4 hectares. The animals are bred on fiscal grounds, no fences, no fixed limits of grazing areas, or directly the owner does not own land. The type of farming is extensive with enclose night. The averaged, of goats, sheep and dogs by GFU is 82, 9±57, 1, 5, 3±4, 7, 4, 9±2, 5 respectively. The unique practice sanitary, which is performed on a routine basis, is deworming, against gastrointestinal nematodes and or *Fasciola hepatica* 1 or 2 times per year. 33 feces samples, of GFU, were collected to be analyzed by the technique of COPROELISA. 55% of the GFU had dog's positives, and 45.4% of the total of sampled dogs was positive for *E. granulosus* sl. 55% of owners not deworming dogs, 20% did not remember and 25% dewormer ever with praziquantel. In this first study, it can be seen the need to implement control measures to prevent the spread of this parasites.

CONTROL OF ECHINOCOCCOSIS IN SICHUAN PROVINCE OF CHINA

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Background: echinococcosis is mainly prevalent in 35 counties in 4 prefectures (cities) of Ganzi, Aba, Liangshan and Ya 'an in sichuan province. According to a national epidemiological survey of echinococcosis in 2012, the average prevalence rate of echinococcosis among the population in Sichuan is 1.08%, Ganzi prefecture is the most serious with the prevalence rate of 1.86%, and Shiqu county is as high as 12.09%, which is rare in the world. Objective: to establish and improve the monitoring system and control echinococcosis. Methods: adopt the comprehensive prevention and control strategy of "control the transmission source, intermediate host prevention and control, and patient screening and treatment", and carry out annual monitoring on the prevalence of cattle and sheep in each epidemic county and the infection of dogs in each epidemic township. Results: between 2011 and 2018, the detection rate of echinococcosis patients decreased from 0.18% to 0.04%, the infection rate of *Echinococcus* spp. in dogs decreased from 19.22% to 5.05%, and the prevalence rate of echinococcosis in livestock decreased from 6.38% to 5.07%. Discussion/conclusion: while making achievements, we are soberly aware that the situation of prevention and control of echinococcosis in Sichuan province is still very serious. In the next step, we will continue to increase support for the prevention and control of echinococcosis, fully implement comprehensive prevention and control measures to curb the epidemic situation of echinococcosis.

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ACHIEVEMENTS IN THE CONTROL OF CYSTIC ECHINOCOCCOSIS IN URUGUAY  
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**Introduction:** Cystic echinococcosis is a parasitic zoonosis caused by *Echinococcus granulosus*, cestode of the Taenidae family. In Uruguay, its life cycle involves canines, sheep and cattle, it is a public health problem and also generates economic repercussions on livestock production. Recently, it was included in the WHO list of neglected diseases, which implied the execution of an action plan for the elimination, 2016–2022. Only Uruguay and China have control programs with national coverage. In our country, the program is the responsibility of the National Zoonoses Commission (CNZ). Our goal is to present the results achieved in these 15 years of uninterrupted work.

**Methods and materials:** The strategy of the CNZ Control Program focuses mainly on a triad consisting of canine dosing, ultrasound diagnosis in humans and a strong component of IEC (information, extension and communication). Dogs are deworming with praziquantel every 30 days; additionally, the canine population is controlled by sterilization. Different evaluation strategies are used, depending on the host; in humans it is performed by ultrasonography studies; in canines by coproELISA; while in cattle is measured in slaughterhouses. Knowledge surveys and habits related to the parasitic cycle are also carried out.

**Results.** From the changes made in the CNZ Control Program, in 2005, it has been observed a marked reduction in the percentage of rural settlements with parasite dogs, from 6.4 to 2.0 (preliminary data); also in cattle (from 11.0 to 3.0) and sheep (7.8 to 1.0). Relating with humans, the rate of cystic images by ultrasonography in 2008 was 0.065 per 100,000 inhabitants. In recent years, ultrasonographic surveillance has intensified in children aged 6 to 14, with no compatible images found since 2015.

**Conclusions:** Analyzing the indicators presented, the effectiveness of the actions of control and surveillance carried out by the National Zoonoses Commission is demonstrated.

SEROPREVALENCE OF FASCIOSIS, HYDATIDOSIS AND CISTICERCOSIS IN MINORS IN  
THREE HOMES OF THE PUBLIC BENEFIT SOCIETY OF CUSCO

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**Objective:** Determine through immunological tests the seroprevalence of fasciolosis, hydatidosis and cysticercosis in the population of minors in charge of the Public Beneficence of the city of Cusco. **Materials and methods:** Cross-sectional study, carried out by seroimmunological tests of ELISA and Immunoblot. **Results:** We evaluated 68 patients, all-underage, 63.24% were female, 62% belonged to the age group of 13 to 17 years and 91.18% came from the department of Cusco. For fasciolosis, 19 positive serums were found by the ELISA test and 06 was confirmed with Immunoblot. Watercress consumption was the highest risk factor for contracting *F. hepatic* finding an OR = 128. In the case of hydatidosis, 19 positive serums were found using the ELISA test and 03 was confirmed with Immunoblot. It was defined that feeding dogs with raw viscera is a very high risk factor for contracting *E. granulosus*, finding an OR = 140. For cysticercosis, 02 positive serums were found by the ELISA test with no confirmation by immunoblot. **Conclusion:** A seroprevalence of 8.82% was found for fasciolosis; 4.41% for hydatidosis and 0% for cysticercosis.

**Keywords:** Fasciolosis; Hydatidosis; Immunology.

INTRACRANIAL CYSTIC EQUINOCOCCOSIS IN CHILDREN OF THE PERUVIAN ANDES:  
REPORT OF NINE CASES ON SURGICAL TECHNIQUE.

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**INTRODUCTION:** Hydatidosis is a chronic, zoonotic, helminthic and endemic disease. Intracranial presentation is common in pediatric age. The treatment of intracerebral cystic echinococcosis is mainly surgical. The objective is complete and unbroken removal, which is why the description of a combined surgical technique based on microsurgery and the Dowling-Orlando technique is proposed. **MATERIAL AND METHOD.** The method used was descriptive and retrospective. Between the period 1998-2019, 9 cases of children diagnosed with intracerebral cystic hydatidosis that were surgically operated at the National Institute of Child Health were included. The place of origin, the clinical manifestations, the neuroimaging findings, the surgical technique performed and the morbidity and mortality associated with the operative technique were recorded. **RESULTS.** The 9 patients studied, 6 (66.6%) were male and 3 (33.3%) female. The average age was 8 years. 8 (88.8%) patients came from the province. The average illness time was 5 months. The most frequent clinical manifestations were intracranial hypertension (100%), focal neurological deficit (66.6%) and convulsive syndrome (25%). The hydatid cyst was located in the cerebral parenchyma in 8 patients (88.8%) and in 1 patient (11.2%) it was located in the posterior fossa. In 7 cases a combination of microsurgery techniques and the technique of Dowling and Orlando was used. The results of the surgical treatment were good in 88.8% (8) leaving discharge in good clinical conditions. An operated patient died. **DISCUSSION AND CONCLUSION.** The best surgical approach for intracerebral cystic echinococcosis should be based on the size and location of the cyst, and its relationship with vascular and neural structures. The combined use of microsurgery and Dowling and Orlando techniques in the excision of the intracerebral hydatid cyst has allowed us to obtain good surgical results for our patients.

**Key words:** cystic echinococcosis, neurosurgery, microsurgery.

PROTEOMIC CHARACTERIZATION OF THE 21-31 kDa ANTIGENIC FRACTION OF LIQUID  
HYDATID CYST OF *Echinococcus granulosus*

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*Echinococcus granulosus* is a cestode which its larval form causes Cystic Echinococcosis (CE). This disease is a parasitic zoonosis with worldwide distribution, which affects humans and cattle. Liquid hydatid cyst (LH) from *E. granulosus*, is used as an antigen in diagnostic tests. LH is a complex mixture that contains both parasite and host proteins, which causes sensitivity and specificity problems. In order to identify parasite proteins and generate recombinant proteins that improve and facilitate diagnosis in humans, a 21-31 kDa protein fraction of LH was purified, by precipitation with ammonium sulfate and using ultrafiltration and electroelution methods. The purified

fraction was analyzed by Mass Spectrometry (MS) and bioinformatic analysis. The genes of the identified proteins were amplified by PCR, cloned into a plasmid which was used to transform the yeast *Pichia pastoris*. The recombinant protein generated was evaluated by immunoblot. The result of the purification of the 21 to 31 kDa fraction of LH showed a single band. MS analysis of this fraction identified a new parasite protein among several host proteins. The new protein corresponds to a tegumentary protein called Neurogenic Locus Notch. Due to the large size of the gene that encodes this protein, three epitopic regions were identified and selected, which were amplified by PCR and cloned into the plasmid to transform the yeast *Pichia pastoris*. The evaluation of the antigenic capacity of the expressed and purified recombinant protein, proved to be specific when faced with sera from patients with CE. The Notch recombinant protein is 18kDa in size and has diagnostic value for the disease.

Keywords: Proteomic, *Echinococcus granulosus*, Recombinant protein

### HYDATIDOSIS OF THE SUBMANDIBULAR GLAND IN CHILDREN.CASE REPORT

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**INTRODUCTION.** Cystic echinococcosis or hydatidosis is a zoonosis produced by the *Echinococcus granulosus* cestode of high prevalence in Peru, central Andean region (Cerro de Pasco, Junín, Huancavelica), with slow growth in humans, can be asymptomatic. In Peru, cumulative incidence rates fluctuate between 39 and 79 cases per 100,000 inhabitants. The hydatid cyst develops mainly in the liver and lung, and 5% in other organs, due to the blood route taken by the hexacanth embryo, which, after crossing the small intestine, reaches the liver and then the lungs to through the pulmonary artery. **OBJECTIVE.** Present an unusual case of hydatidosis in the submandibular gland. **MATERIAL AND METHOD.** A case of hydatidosis of right submandibular location is reported in a 12-year-old girl, 3 months old, born in Andahuaylas, illness time of 04 years. Soft tissue ultrasound of head and neck: right submandibular collection. Complete abdominal ultrasound and frontal chest x-ray: normal. Soft tissue ultrasound of the neck: right submandibular gland cyst. Serology: negative. Exeresis of the lesion was performed with a good evolution. Pathological anatomy: study of the submandibular gland. **RESULTS.** Pathological anatomy: right submandibular gland: brownish tissue formation of 4.2x3, 3x1.1 cm. smooth surface, reticent consistency, weight 7 gr. The laminations identify cystic formation of partially defined edges of 2.2x2x0.8 cm. in contact with the edge of the section, whitish internal surface, folded, mucinous content. **Diagnosis:** Anhist membranes of viable hydatid cyst. **DISCUSSION AND CONCLUSION.**The hydatid cyst in the submandibular gland in a pediatric patient is highly atypical. The clinical picture is nonspecific. The diagnosis is by pathological anatomical study.

Key words: echinococcosis, submandibular Gland, children

### STUDY OF 3.0 T MR DIFFUSION WEIGHTED IMAGING IN HEPATIC CYSTIC ECHINOCOCCOSIS

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**Objective** To explore the relationship between apparent diffusion coefficient (ADC) value and the classification of different hepatic cystic echinococcosis (HCE) and to evaluate the biological activity of the lesions. **Methods** By setting different  $b$  values ( $b=100, 300, 600, 800, 1000, 1200$  and  $2000\text{s/mm}^2$ ), 64 patients with HCE were scanned with 3.0T MR diffusion weighted imaging (DWI). Respectively, the DWI manifestations and ADC values of the same lesion were analyzed under different and the same  $b$  value. Patients were scanned by DWI before and after the therapy of medication or microwave, and the ADC values were measured. **Results** Lesions were showed better when  $b=800\text{s/mm}^2$  was performed. HCE classification: there were 34 cases in CE1, 34 cases in CE2, 9 cases in CE3, 31 cases in CE4 and 8 cases in CE5. There were significant differences of the ADC values among groups of CE1, CE2, CE4 and CE5 ( $P<0.05$ ). ADC values of CE2, CE4 and CE5 were significantly different ( $P<0.05$ ). There was no significant difference of ADC values between CE3 and the other four types ( $P>0.05$ ). The ADC values of lesions decreased after 3 months of therapy of medication or microwave. The difference was statistically significant ( $P<0.05$ ). **Conclusions** MR DWI sequence and ADC value are helpful in the diagnosis and classification of HCE. The ADC values of five types of HCE lesions have decreased with the decrease of lesion activity, and it is useful to evaluate the efficacy of medication or microwave therapy.

**Keywords:** hepatic cystic echinococcosis; diffusion weighted imaging

#### RE-EVALUATION OF CLASSIFICATION OF HEPATIC CYSTIC ECHINOCOCCOSIS BY MULTIMODAL IMAGING

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**Objective** The classification of the WHO--OIE hepatic cystic echinococcosis was mainly divided into CE five types by means of B-ultrasound in 2001. In this study, the classification of HCE was re-evaluated by multi-modality imaging techniques such as B-ultrasound, CT and MRI. And clarify the advantages of all kinds of imaging technology in HCE classification. **Methods** Cystic lesions of the liver are detected by means of diagnostic imaging methods, such as ultrasonography, magnetic resonance imaging (MRI), and computed tomography. 1000 cases suffered from HCE underwent B-scan ultrasound and CT simultaneously, 450 cases underwent MRI, and 300 cases underwent CT and MRI simultaneously. **Results** A total of 1754 lesions of hepatic cystic echinococcosis were collected. According to the classification which was proposed by the WHO Working Group on echinococcosis for cystic echinococcosis, the lesions were divided into five types: CE I (178, 10%), CE II (664, 38%), CE III (554, 32%), CE IV (212, 12%) and CE V (146, 8%). The detection rates of calcification of the cyst wall, internal septa, floating membranes and daughter vesicles by CT were significantly higher than those by US ( $P<0.05$ ). The detection rate of calcification by CT was significantly higher than those by US and MRI ( $P<0.05$ ). MR is the best imaging procedure to demonstrate a cystic component and evaluate the activity of cystic echinococcosis. It helps to determine vascular or biliary tree involvement, as well as extrahepatic extension. The specificity of MRI was significantly higher than US and CT in the differential diagnosis of CE I, CE II and CE III. **Conclusion** The use of multimodal imaging technology is more accurate in the classification of HCE. It is helpful to judge the activity of HCE lesions and to choose the treatment methods.

**Key words** Hepatic cystic echinococcosis; Multimodal imaging; Ultrasonography; Computed tomography; Magnetic resonance imaging; Classification

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CASE REPORT: MUSCULOSKELETAL HYDATID DISEASE TREATED WITH ALBENDAZOLE  
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Cystic echinococcosis caused by *Echinococcus granulosus*, is an important problem of Public Health in Uruguay.

Musculoskeletal hydatid disease is uncommon but severe, usually late diagnosis and the treatment includes surgical resection and prolonged pharmacological approach. We report a case of a 79 years old female, from Soriano - Uruguay. She was derived to the Department of Parasitology and Mycology in 2003, due to a musculoskeletal cystic echinococcosis of 38 years evolution, located in the right hemipelvis and femur and adductor mayor's lodge. She was surgically treated in 1974 and subsequently remained in traumatological follow-up, due to fistulization of the skin in the right coxo-femoral region with exit of daughter vesicles of *Echinococcus granulosus* accompanied by intense pain that limited her walking. She was treated with cycles of Albendazole over 9 years, with clinical, imaging and Hemogram, Functional and Hepatic Enzymogram monitoring. Her painful symptoms disappeared as well as the exit of parasitic elements through the skin with spontaneous closure of the fistulous tract, thus improving her quality of life. Epigastric pain and heartburn were observed during the first two cycles, improving with inhibitors of the proton pump. Echinococcosis is preventable with diagnosis and early and appropriate surgical treatment which improve survival. In cases when surgery is not possible or when complete resection of the cyst is not achieved, medical treatment, with Albendazole alone or in combination with Praziquantel, is promising. It is important to continue investigating to determine what should be the optimal duration of medical treatment, to evaluate the possibility of using new drugs, or the instillation of substances during the surgical act to prevent recurrences.

STUDY ON THE RELATED EXPENSES OF HYDATIDOSIS SURGICAL PATIENTS TREATED  
IN COUNTY-LEVEL DESIGNATED HOSPITALS OF XINJIANG UYGUR AUTONOMOUS  
REGION FROM 2014 TO 2017

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Objective: To explore the hospitalization expenses of patients with hepatic cystic echinococcosis (HCE) in county hospitals in Xinjiang, and to analyze the implementation of the state subsidy policy. Method. Describe the constituent ratio, mean and median descriptions of hospitalization expenses of surgical treatment patients with HCE in county hospitals of Xinjiang from 2014 to 2017. Analysis the influencing factors of hospitalization expenses by single factor and logistic stepwise regression analysis. Result A total of 841 HCE surgical patients were investigated. The total hospitalization cost was 10 278 126.47 yuan, 12 221.32 yuan per capita, with a median of 11 500.66 yuan. Among them, drug expenses accounted for the largest proportion (32.48%). The significant influencing factors of hospitalization expenses were the length of hospitalization days, the availability of medical insurance and the number of lesions. The total amount of the state subsidy is 300, 6318.11 yuan, with an average of 3,574.69 yuan per capita, with a median of 3,200.62 yuan, with the largest number of people from 2000 to 4,000 yuan in each year. After medical insurance and state subsidy, 772 patients paid 0 yuan, accounting for 91.80% of the total number. 69 cases still need to pay part of the hospitalization expenses by themselves, and the largest number of people pay less than

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2000 yuan each year. Conclusion The national economic subsidy policy can basically meet the needs of HCE patients treated in county hospitals. Hospitals should pay attention to adjusting the proportion of drugs, improving technical level and controlling days, so as to control the total cost of hospitalization.

Keywords: County hospital; Hydatid disease; Related costs; Subsidy

#### ANALYSIS OF QUALITY OF LIFE AFTER END-STAGE HEPATIC BUBBLE HYDATID DISEASE

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**Objective:** To analyze the quality of life of end-stage hepatic bubble echinococcosis after operation, according to the comparison and analysis before and after operation in March and June, to understand the quality of life of end-stage hepatic. **Methods:** The preoperative postoperative situation of 82 patients with hepatic bubble echinococcosis treated by autologous liver transplantation in the first affiliated hospital of Xinjiang Medical University since May 31, 2015 to May 31, 2018 was compared and analyzed, and the appropriate quality of life scale was selected, and the application survey was. **Results:** There were statistical differences ( $P < 0.05$ ) between March and June after liver transplantation compared with preoperative, and there was no significant statistical significance compared with June after operation in March. **Conclusion:** By studying the quality of life before and after autologous liver transplantation of end-stage hepatic bubble echinococcosis, it is found that the overall quality of life of patients after operation is significantly higher than before operation, and the current autologous liver

#### ECHINOCOCCUS GRANULOSUS: APPROACH ON ENVIRONMENTAL IMPACT.

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Cystic Echinococcosis is a parasitic zoonosis that is transmitted by environmental contamination with *Echinococcus granulosus* eggs. The domestic cycle establishes by contact of the definitive guests with the parasitized viscera of the intermediate hosts slaughtered (zoonotic cycle) and between the definitive hosts and the environment (environmental cycle), being the man an intermediate host of the environmental cycle. In order to gather information about the epidemiological status regarding *Echinococcus granulosus* of the canines of the municipality of Pampa del Indio and of the Curishi settlement community, El Descanso, located on the outskirts of the city of J.J. Castelli, both from the Province of Chaco, Argentina, for future actions in what this disease implies. **Materials and methods:** 79 samples of dry fecal matter (MFS) were collected from canines in the peridomicil of rural dwellings in 7 places of Pampa del Indio and 163 samples of El Descanso settlement. The samples were processed individually by conventional parasitological methods and by the application of the specific PCR technique for *Echinococcus granulosus*, amplifying a partial sequence of the mitochondrial gene of subunit 1 of cytochrome c oxidase, which aims to demonstrate specific DNA sequences of the parasite. **Results:** total samples analyzed (242), 6 positive samples were detected by microscopy for eggs of *Taenia* sp., Of these samples, one was positive by specific PCR for Eg. and of the 236 negative samples by microscopy, 10 samples were positive by specific PCR for *Echinococcus granulosus*. **Conclusions:** The

presence of eggs of *Taenia* spp. and subsequent characterization as eggs of *Echinococcus granulosus*, is evidence that there are risky habits of local people, such as feeding dogs with raw viscera, increasing the risk of infection by *E. granulosus* for people who inhabit these areas.

Palabras clave: *Echinococcus* egg diagnosis

**MULTI-CENTRE STUDY ON *ECHINOCOCCUS MULTILOCULARIS* AND *ECHINOCOCCUS GRANULOSUS S.L.* IN EUROPE: DEVELOPMENT AND HARMONIZATION OF**

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**INTRODUCTION:** MEmE is a multicentre collaborative international project (2020–2022) composed by 17 partners from 13 countries in Europe and Asia. MEmE is aiming to fill the research gaps highlighted by international agencies for the detection and control of cystic and alveolar echinococcosis (CE and AE). **MATERIALS and METHODS:** MEmE, expanding the results of previous European projects, will focus on both the standardization, harmonization and validation of existing parasitological and molecular methods, and the development and comparative assessment of innovative molecular tools and biomarkers to detect *Echinococcus multilocularis* (Em) and *Echinococcus granulosus s.l.* (Eg) in the food chain. A biomarker discovery task will also focus on the proteomic analysis of exosomes in sheep plasma, to develop innovative tools for the detection of CE the natural intermediate host. Production of epidemiological data on the presence of Em/Eg eggs in the food chain will focus on vegetables for human consumption as well as dogs' faeces in selected endemic countries. Moreover, food source-targeted questionnaires will be developed and administered to a sample of patients with CE, in selected hospitals, including those registered in the European Register of CE (ERCE), and AE and matched controls, to advance our knowledge on food-related risk factors for human infection. Organization and delivery of parasitological and molecular Proficiency Testing Schemes on the validated techniques will be organized. **DISCUSSION/CONCLUSIONS:** Altogether, MEmE will provide a comprehensive set of relevant integrative activities (including development and validation of protocols, collection of biological material, capacity building, and epidemiological investigations). These will allow partner organisations to harmonize procedures, improve detection of Eg and Em, and define control strategies based on the occurrence of these pathogens in the food chain and the relative importance of their food-borne transmission. This work is funded by the European Commission (Horizon 2020) under One Health European Joint Programme (<https://onehealthjp.eu/>).

**Keywords:** *Echinococcus* spp., detection methods, epidemiology

**ECHINOCOCCUS GRANULOSUS IN AUSTRALIAN WILDLIFE: A RESERVOIR PERPETUATING TRANSMISSION OF INFECTION TO LIVESTOCK.**

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*Echinococcus granulosus* was introduced into Australia in the late 1700s and early 1800s, during settlement, with domestic dogs and livestock (mainly sheep). Domestic transmission soon became established between domestic dogs and sheep and before too long hydatid disease became a major public health issue in all the sheep rearing areas of eastern and southern Australia. Australian wildlife consisted of marsupial herbivores and carnivores that had not evolved with *E. granulosus* and, by coincidence, also a placental canid, the dingo (*Canis lupus dingo*) that had been introduced from South East Asia a few thousand years before and had also not been exposed to *E. granulosus*. Dingoes and macropod marsupials proved to be highly susceptible to infection with *E. granulosus* and a wildlife reservoir became established through predation of macropod marsupials by dingoes. Since the recent discovery of the highly efficient cestocidal drug, praziquantel, and the development of nutritious, highly palatable dry dog food, *E. granulosus* now occurs less commonly in rural dogs and hydatid infection in sheep has also become less common in most Australian sheep rearing regions. However, hydatid disease occurs commonly in sheep on farms that abut national parks, state forests or areas of unowned land where dingoes live. Dingoes move across the farms, often also killing sheep, but importantly defecating and releasing eggs of *E. granulosus* into the environment to be spread by a range of agents such as wind, rain and coprophagous flies. Cattle remain commonly infected with *E. granulosus* as a result of being commonly grazed on bush pasture inhabited by dingoes. Also in Australia are populations of domestic animals (donkeys, camels, goats and pigs) that have become feral and are now living as wild animals and their role in transmission will be discussed.

## 30 DE OCTUBRE

### INTERNATIONAL EVALUATION OF VASCULAR / BILIAR INVOLVEMENT OF HEPATIC LESIONS AND EXTRAHEPATIC DISEASE MANIFESTATION DUE TO HEPATIC ALVEOLAR ECHINOCOCCOSIS ACCORDING TO EMUC-CT CLASSIFICATION: A MULTICENTRE COHORT STUDY

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Introduction: The main endemic areas of alveolar echinococcosis (AE) are in Central Europe and Western China. Aim of this study was to classify a great collective of German, French, and Chinese cases of hepatic alveolar echinococcosis (AE) in accordance with the *Echinococcus multilocularis* Ulm classification for computed tomography (EMUC-CT)

and to evaluate the vascular / biliar involvement and the extrahepatic manifestations. Material and Methods: Fifty contrast-enhanced CT scans of patients with hepatic AE were evaluated from each localities (n=200): Xining, Urumqi, China; Ulm, Germany; Besançon, France (XUUB). Liver lesions were classified according to EMUC-CT. Cases were evaluated regarding vascular / biliar involvement of the liver lesions and the extrahepatic manifestations. Results: Extrahepatic AE manifestations were significantly more frequent in China than in Europe (p=0.0091). EMUC-CT type IV and V, unlike types I, II and III, show no cases of extrahepatic disease manifestation. Type III is most frequently associated with extrahepatic manifestation (22.22%). A significant relationship exists between the presence of an extrahepatic disease manifestation and the size of the liver lesion overall (p=0.0075) and for China (p=0.0251). Vascular / biliary structures are significantly more frequently infiltrated in China than in Europe (p<0.0001). A significant correlation exists between infiltration of vascular / biliary structures and lesion size. Different types lead to varying frequency of vascular / biliary infiltration with a minimum for type IV (5.88%) and a maximum for type III (100%). Comparing the groups without and with vascular / biliary involvement with regard to extrahepatic disease manifestations for the overall collective, it is interesting to note that there is no significance for this. Discussion/Conclusion: The results can provide important information on the possible behaviour of the disease during initial manifestation and progression. Extrahepatic AE manifestations do not appear to be metastases originating from liver lesion, but also primary manifestations.

Keywords: Alveolar echinococcosis; intercontinental; Echinococcosis Multilocularis Ulm Classification for Computed Tomography (EMUC-CT); vascular / biliar involvement; extrahepatic manifestations.

## A STUDY OF MAGNETIC RESONANCE IMAGING OF IRON DEPOSITION IN HEPATIC ALVEOLAR ECHINOCOCCOSIS

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**Objective** In view of the invasive growth characteristics of hepatic alveolar echinococcosis, the destruction of blood vessels and bile ducts is obvious in the middle and late stages, and the high degree of liver damage, the serum ferritin is significantly elevated in most patients, the study is aimed to analyze the liver iron deposition and liver function in hepatic alveolar echinococcosis(HAE) patients using MRI together with serum ferritin. **Methods** 96 HAE patients and 30 normal volunteers were all scanned with 1.5T MR, and relevant data of laboratory inspections were collected. The signal intensity ratios of liver to the paraspinous muscles (L/M-SIR) were measured on TSE T2WI-SPAIR. The correlation between L/M-SIR and serum ferritin, L/M-SIR and liver function were analyzed. Futhermore, 96 HAE patients were divided into normal serum ferritin group[(4.63-204) $\mu$ g/L, 30 patients] and increased serum ferritin group(>204 $\mu$ g/L, 66 patients), and laboratory markers determining the liver function were analyzed. **Results** The L/M-SIR of HAE group and normal group were 1.95 $\pm$ 1.30 and 2.41 $\pm$ 0.53 seperately, which indicated significant differences between groups. Moreover, there was a negative correlation between L/M-SIR and serum ferritin( $r=-0.44$ ,  $P<0.01$ ) in HAE group. Significant differences( $P<0.05$ ) of liver function data were found between normal serum ferritin group and increased serum ferritin group in HAE patients. **Conclusion** Iron deposition is common in HAE, serum ferritin can be used to detect liver iron deposition of HAE patients, and the degree of liver damage is related to iron deposition. MRI, a non-invasive method, is efficient to assess liver iron deposition in HAE.

Key words : Hepatic alveolar echinococcosis; Iron deposition; Serum ferritin; Magnetic resonance imaging

## THE VALUE OF "ONE-STOP" COMPREHENSIVE MR EVALUATION IN THE DIAGNOSIS OF HEPATIC ALVEOLAR ECHINOCOCCOSIS

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**Objective** Hepatic alveolar echinococcosis (HAE) is demonstrated as a map-like heterogenous mass and aggressive. The lesions may compress or destroy the hepatic vessels or bile ducts. To investigate the value of "one-stop" comprehensive MR evaluation of HAE for clinical diagnosis and surgical decision . **Methods** 98 patients with HAE from November 2011 to June 2017 in the hospital were underwent "one-stop" MRI before operation . MRI scan was performed including plain scan , diffusion weighted imaging(  $b=0 \text{ mm/ s}^2$  ,  $b=800 \text{ mm/ s}^2$  ) , enhanced scanning with conventional 4 phase and 3D magnetic resonance imaging cholangiography (MRCP). **Results** (1)There were 98 patients with a total 145 lesions of HAE . Based on imaging findings the HAE lesions were divided into five types: solid type (Type I , 53/145) , liquefied necrosis type in center area (Type II , 65/145 ) , mixed type (Type III 15/145) , multiple nodular type (Type IV , 9/145) and calcified type (Type V , 3/145 ) . (2)167 branches of the intrahepatic vein , 65 branches of the hepatic artery and 87 branches of portal vein were involved . (3) MRCP could clearly show the lesions that located at or extended to hepatic hilum caused dilatation of intrahepatic biliary ducts (45/145) and bile duct stenosis location and degree . (4) The ADC values of the solid part ,the surrounding liver tissue ,and the peripheral zone decreased in turns [ $(1.54\pm 0.13)\times 10^{-3} \text{ mm}^2/\text{s}$  ,  $(1.39\pm 0.17)\times 10^{-3} \text{ mm}^2/\text{s}$  ,  $(1.01\pm 0.13)\times 10^{-3} \text{ mm}^2/\text{s}$ ] , showing significant differences between each two values ( $P<0.05$ ) . (5) Compared to T2W\_SPAIR and DWI, the MRCP sequence has obvious advantages for the detection and display of microcysts. **Conclusion** "One-stop" comprehensive MR evaluation can successfully delineate the involvement of hepatic vascular and biliary system and can be used in the preoperation evaluation of HAE as routine examination .

**Key words** hepatic alveolar echinococcosis ; magnetic resonance imaging ;magnetic resonance angiography ; magnetic resonance cholangiography

## REGULATORY EFFECT AND MECHANISM OF CD4+CD25+CD127LOW/ FOXP3 TREG CELLS IN ALVEOLAR ECHINOCOCCOSIS

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**Objective:** To investigate the expression of Foxp3 in CD4+CD25+CD127lowTreg cells and to study the mechanism of CD4+CD25+CD127low/ Foxp3 Treg cells in alveolar echinococcosis. **Methods:** The mice were divided into alveolar echinococcosis group and healthy control group. The percentage of CD4+CD25+CD127lowTreg cells and CD4+CD25+Foxp3 Treg cells and the expressions of Foxp3 in CD4+CD25+CD127lowTreg cells were detected by flow cytometry. The levels of IL-10 and TGF- $\beta$ 1 in serum were detected by ELISA. The relationship between CD4+CD25+CD127low/ Foxp3 Treg cells and lesion size was analyzed by Pearson Correlation Analysis. **Results:** 1. The percentage of CD4+CD25+CD127lowTreg cells in the healthy control group was  $9.04\pm 2.78$ , while it was  $14.91\pm 4.21$  in the alveolar echinococcosis groups ( $P<0.004$ ) . 2. The percentage of CD4+CD25+Foxp3 Treg cells in the healthy control group was  $0.76\pm 0.73$  and  $4.21\pm 2.08$  in

the alveolar echinococcosis group ( $P < 0.000$ ). 3. Foxp3 in CD4+CD25+CD127lowTreg cells was 70.7% in healthy control group and 67.7% in the alveolar echinococcosis group. 4. The expression of CD4+CD25+CD127lowTreg cells in alveolar echinococcosis group was positively correlated with the size of the lesions ( $r=0.894$ ,  $P=0.01$ ). The expression of CD4+CD25+Foxp3 Treg cells in alveolar echinococcosis group was positively correlated with the size of the lesions ( $r=0.714$ ,  $P=0.031$ ). 5. The level of IL-10 in the healthy control group was  $533.82 \pm 55.82$ , while it was  $1946.80 \pm 260.89$  in the alveolar echinococcosis group ( $P < 0.000$ ). The level of TGF- $\beta$ 1 in the healthy control group was  $223.90 \pm 56.34$ , while it was  $321.35 \pm 36.44$  in the alveolar echinococcosis group ( $P < 0.01$ ). Conclusion: 1. Echinococcus multilocularis infection can stimulate the proliferation of CD4+CD25+CD127low/ Foxp3 Treg cells in mice, and CD4+CD25+CD127lowTreg cells express Foxp3 highly, which can be used as surface markers of Treg cells in alveolar echinococcosis; 2. The expression of CD4+CD25+CD127low/ Foxp3 Treg cells was positively correlated with the size of the lesions. Immunosuppressive cytokines (IL-10, TGF- $\beta$ 1) secreted by Treg cells from the infected mouse were highly expressed, which indicated CD4+CD25+CD127low/Foxp3 Treg cells can promote the development of alveolar echinococcosis, and the mechanism may be related to the inhibition of the body's immune function.

#### SALVAGE TREATMENT WITH AMPHOTERICIN B FOR ALVEOLAR ECHINOCOCCOSIS CAN INDUCE LONG-TERM DISEASE CONTROL

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Background: Curative resection is the treatment of choice for alveolar echinococcosis (AE) with circumscribed manifestation. However, complete resection is frequently impracticable due to extended involvement of critical anatomic structures. Benzimidazole treatment (BMZT) is chosen in such cases, but may be limited due to inefficacy or intolerance. Amongst other experimental approaches, amphotericin b treatment (AMBT) has been used with success in desperate situations. Methods: We here report long-term follow-up data from 4 patients with AMBT at our center who were diagnosed with unresectable AE with BMZT intolerance or resistance. Initial AMBT loading phase of 2 weeks was followed by 1-3 applications per week, depending on response. Close monitoring was conducted for the first month and after that every 3 months. Results: All patients had complex disease manifestation with need for repeated surgical interventions due to complications. Three patients exhibited disseminated disease with pulmonary, cardiac, vertebral and cerebral manifestations. Patients showed

intolerance (n=2) and progression (n=2) under BMZT. Age at diagnosis ranged from 42-58 years and AMBT was initiated 10-25 years after diagnosis and a median of 3 previous lines of systemic therapy. With AMBT, stable disease (SD) was achieved for 2 months and 13 years and AMBT is currently ongoing in these patients. Two patients died after achieving SD for 3 years and a partial remission for 17 months due to progress later during the disease course. Responses were confirmed by PET-CT scan, showing stable or regressive metabolic activity or lesion size. Further, clinical evidence of response was documented with healing AE associated cutaneous fistulae and regression of abdominal discomfort. Side effects under AMBT were mostly manageable, however deteriorating renal function after 3 years prompted the halt of AMBT in 1 patient. Discussion: Salvage AMBT is a highly valuable treatment option and can induce persistent disease control.

#### EVALUATION THE VIABILITY OF ALVEOLAR ECHINOCOCCOSIS: A COMPARISON BETWEEN [18F] FLUORODEOXYGLUCOSE POSITRON-EMISSION TOMOGRAPHY AND SPECTRAL CT

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**Purpose:** To assess the iodine concentration of hepatic alveolar echinococcosis(HAE) using spectral computed tomography (CT)with comparison of [18F]fluorodeoxyglucose positronemission tomography (FDG-PET),to explore the value of spectral CT for evaluation of HAE viability. **Materials and methods:** 18 patients with histologically confirmed or clinically proved HAE underwent spectral CT and FDG-PET examinations. After three-phase(arterial phase (AP), portal venous phase (PVP) and venous phase(VP)) scan, the raw data transfer to advanced 4.6version work station for measurement .The quantitative iodine-based material decomposition images and optimal monochromatic image of spectral CT were reconstructed and iodine concentration (IC) was measured in different organizational structures. Iodine results of different phases and different tissues were compared with Rank test. The correlation of iodine value and PET SUV were performed with Spearman method. **Results:** FDG-PET identified increased metabolic activity in the corresponding lesions in 13 patients (13/18, 72.2%).The iodine concentration in marginal zone of lesion were significantly higher than in solid component of lesion and normal liver parenchyma during PVP and VP . Positive FDG-PET findings with "hot spot" were also positive at spectral CT images which displayed as continuous or discontinuous ring-like enhancement with the 5~10mm thickness.The iodine value of hydatid tissues showed no statistically significant difference ( $P>0.05$ ). The iodine value of edge tissue of the lesion and normal liver and iodine value of normal liver tissues showed statistically significant difference ( $P <0.001$ ).There was correlation between IC and SUV in marginal zone of HAE lesion, and it was highest during PVP ( $r = 0.873$ ,  $p <0.001$ ).There was low between CT values and SUV. **Conclusion:** There were good correlation between spectral CT and FDG-PTE . Spectral CT could be recommended as a more practical tool in the clinical routine.

#### MRI CHARACTERISTICS AND TREATMENT EVALUATION OF BILE DUCT INVASION IN ADVANCED HEPATIC ALVEOLAR ECHINOCOCCOSIS

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Object Advanced hepatic alveolar echinococcosis is prone to invade intrahepatic bile duct. Obstructive jaundice is the main clinical manifestation and also an important restriction factor to reduce the surgical resection rate of echinococcosis patients. MRI was used to identify the features of intrahepatic bile duct invasion in HAE, and the therapeutic effects after PTC and ERCP were evaluated. Methods Before treatment, 35 patients with advanced HAE underwent conventional 1.5T MRI routine scan and magnetic resonance cholangiopancreatography. The scanning parameters were consistent with the image reconstruction method. 35 patients were treated by PTC and ERCP and then evaluated for efficacy. The evaluation content is as follows 1. Anatomical morphological changes of bile duct damage in 35 cases of hepatic alveolar echinococcosis. 2. Chi-square test was used to compare the invasion rates of left hepatic duct, right hepatic duct and common hepatic duct. 3. The jaundice index and liver function index of 35 patients with advanced hepatic alveolar echinococcosis before PTC and ERCP, 1 week after operation and 1 month after operation were compared. Result 1. Anatomical morphological changes of bile duct damage in 35 cases of hepatic alveolar echinococcosis. The morphology of bile duct was not affected in 5 cases, 17 cases of bile duct stenosis with obstruction and dilation, 13 cases of bile duct truncation and obstruction expansion. 2. The total invasion rate of bile duct was 36.6%, The invasion rates of the left hepatic duct, right hepatic duct and common hepatic duct were 26.4%, 41.4% and 42.1%, respectively, with overall  $P < 0.05$ ; Through the consistency test with surgery, the Kappa coefficients of the three groups were 0.602, 0.695 and 0.741, respectively. 3. All 17 cases of PTC were successful, ERCP was performed in 18 cases, of which 5 cases failed and 13 cases succeeded. Jaundice index and liver function index were significantly lower than those before operation one week and one month after operation,  $P > 0.05$ . Conclusion MRI can accurately evaluate the invasion degree of HAE lesion to biliary tract and help clinical selection of treatment methods. PTC and ERCP can effectively reduce jaundice and improve liver function, and can be used as an effective treatment for hepatic alveolar echinococcosis complicated with obstructive jaundice. For patients with ERCP failure, PTC can be selected for treatment.

Keywords Hepatic alveolar echinococcosis; MRI; Bile duct invasion; Endoscopic retrograde cholangiopancreatography; Percutaneous transhepatic cholangial drainage

#### QUANTITATIVE STUDY OF FAT DEPOSITION AND IRON DEPOSITION IN PATIENTS WITH HAE BY HISTO AND Q-DIXON TECHNIQUES ON MRI

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Objective In view of the invasive growth characteristics of hepatic alveolar echinococcosis, the destruction of blood vessels and bile ducts is obvious in the middle and late stages, and the high degree of liver damage, the serum ferritin is significantly elevated in most patients, it is important to analyzed the liver iron deposition and fat deposition in hepatic alveolar echinococcosis (HAE) patients with HISTO and q-DIXON imaging, and correlation analysis with serum ferritin. Methods 15 cases with HAE and 20 healthy volunteers were enrolled in this study. Q-Dixon and Histo sequence were performed on 3.0T MR scanner. The HISTO, could automatically scan the selected region of interest the area under the curve which means the fat fraction was calculated, and draw the curve, and R2 value of the target also can be displayed. The measurement of the region in Dixon imaging of interest should be close to ROI of HISTO. Then, measured the fat fraction and the R2\* value in fat fraction maps and R2\* relaxation maps. Results The R2\* value of HAE group and normal group were  $133.05 \pm 99.74$  and  $88.74 \pm 52.77$ , the R2 value of HAE group and normal group were  $39.98 \pm 8.71$  and  $51.07 \pm 15.94$ . Most patients in the HAE group had higher R2\* value and R2 value than the normal group. Statistically

significant differences in R2\* value and R2 value were found between two groups. A positive correlation was found between R2\* value and serum ferritin (P<0.05, r=0.601) in HAE group. There is no statistically significant differences in R2\* value and R2 value were found in HAE group. A positive correlation was found between R2 value and serum ferritin (P<0.05, r=0.612) in HAE group. The HISTO and Dixon sequences measured liver fat changes in patients without fat deposition. Conclusions Iron deposition is common in HAE, and their fat deposition was basically normal. HISTO and Dixon imaging were sensitive to iron deposition and fat deposition, they can be used as an effective method for non-invasive and quantitative evaluation of HAE iron deposition.

Key words Hepatic alveolar echinococcosis; Iron deposition; Fat deposition; Serum ferritin; Magnetic resonance imaging

#### COMPARED THE AFFECT ABOUT HEPATIC ALVEOLAR ECHINOCOCCOSIS AFTER MICROWAVE ABLATION BY RESOLVE SEQUENCE AND DWI SEQUENCE

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**Objective** To compare the effect about hepatic alveolar echinococcosis after microwave ablation by resolve sequence and DWI sequence with 3.0T MR, Siemens Prisma . **Methods** 50 patients with early hepatic alveolar echinococcosis (HAE) who were given microwave ablation treatment were scanned with 3.0T MR (Siemens Prisma), the sequence scanned including conventional MRI , dynamic-enhance MRI , resolve sequence and DWI sequence of MRI . Those sequences have 2.5x2.5x2.5 mm<sup>3</sup> spatial resolution , 800 s/mm<sup>2</sup> b value, 30 gradient direction, 5 segmentation, 2400ms TR, 52ms TE, 0.34ms Echo spacing, 180 degree Flip angle, 3 Diff. directions, and 919/HZ/Px. Then, to measure and analyze the maximum diameter of the lesion, the edge band of the lesion and the apparent diffusion coefficient (ADCs) of the solid part. **Results** (1) The width of infiltration zone at the edge at resolve sequence and DWI (b=0mm/s<sup>2</sup>, b=800mm/s<sup>2</sup>) were (0.91±0.17)cm, (0.70±0.15)cm, which indicated significant differences between two groups (P<0.05) .The resolve sequence showed statistically significant differences in the thickness of the lesion edge (P<0.05). (2) The evaluation of microscopic necrosis in the lesions was statistically different (P<0.05). **Conclusion** Resolve sequence can significantly improve the image signal-to-noise ratio, reduce geometric deformation and improve spatial resolution. It has a significant value of clinical application.

Key words: Magnetic resonance imaging; Hepatic alveolar echinococcosis; Resolve sequence; DWI sequence; Siemens Prisma

#### THE ASSOCIATION BETWEEN PYROPTOSIS AND HEPATIC ALVEOLAR ECHINOCOCCOSIS : A PRELIMINARY STUDY.

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**Aim:** To explore preliminary the association between the pyroptosis and hepatic alveolar echinococcosis (HAE). **Methods:** 21 cases of normal liver tissue and lesion edge tissue and 20 cases of serum from HAE patients were collected from November 2017 to October 2018 in the Qinghai University Affiliated Hospital. 38 cases of serum samples were collected from healthy volunteers. The key molecules of the pyroptosis signal pathway (caspase-1, caspase-4, caspase-5, GSDMD, IL-18, and IL-1β) were detected by immunohistochemistry and enzyme-linked immunosorbent assay in the lesion edge belt

and the normal liver tissue and serum. Results : Caspase-1, caspase-4, GSDMD, IL-18, and IL-1 $\beta$  were higher in the lesion edge belt tissue than in normal liver tissue. There was no significant statistical difference in serum IL-18 and IL-1 $\beta$ . Conclusion : Pyroptosis maybe occurs in hepatic alveolar echinococcosis and is mainly caused by non-classical pyroptosis mediated by caspase-4.

#### INFLUENCE OF SYSTEMIC CHEMOTHERAPY ON GROWTH OF ALVEOLAR ECHINOCOCCOSIS

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Introduction: Patients suffering from alveolar echinococcosis (AE) might get a malignant tumor additionally. Some of these patients with AE and a concomitant malignant tumor receive systemic chemotherapy. Material and Method: We investigated the growth behavior of AE in these during systemic chemotherapy. Results: We identified 42 patients with malignant disease in addition to a AE. In 6 of out these 42 patients, a systemic chemotherapy was administered. All these patients were on long term anti-helminthic therapy, also during chemotherapy. With respect to alveolar echinococcosis, all these 6 patients showed stable disease according to Response Evaluation Criteria In Solid Tumors (RECIST) during systemic chemotherapy. Four of these patients showed even slightly smaller AE affected liver areas after chemotherapy. Conclusion: Systemic chemotherapy is not associated with a progression of medicamentous treated AE.

#### IMPACT OF LIFE HABITS ON ALBENDAZOLE METABOLISM IN PATIENTS WITH ALVEOLAR ECHINOCOCCOSIS. KEY WORDS: ALVEOLAR ECHINOCOCCOSIS, ALBENDAZOLE, METABOLISM

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Therapeutic drug monitoring offers a personalized approach with dosage adjustments in alveolar echinococcosis (AE) patients under albendazole (ABZ). Many co-medications

may interact with ABZ (1). We describe 3 cases for which life habits led to strong ABZ metabolism interferences. Case 1: a 44-year-old woman was diagnosed for AE in 1993; during a 21-years follow-up she had stable lesions under ABZ 400 mg twice daily, with stable ABZ sulfoxide (ASOX) blood levels within the recommended range. In 2014, low ASOX levels were observed. There were no co-medications or poor observance. ABZ was increased to 600 mg twice daily without impact on blood levels that remained low for 2 years. At the same time, AE lesions progressed. Finally, the patient mentioned regular cannabis consumption since 2014. Cannabis interruption was rapidly followed by increased ASOX levels allowing ABZ dosage reduction. Case 2: a 51-year-old man was diagnosed for AE in 2009. He was a heavy smoker. Under 400mg ABZ twice daily, ASOX levels were low. ABZ dosage was increased under pharmacological surveillance. Finally, 1400 mg/d ABZ were necessary to reach recommended blood level. In 2018, tobacco consumption reduction led to subsequent ASOX levels increase. Case 3: a 64-year-old woman was diagnosed for AE in 2016. Under ABZ 400mg twice daily, ASOX levels were above the therapeutic range, leading to dosage reduction. Under 200mg ABZ twice daily, ASOX levels remained high but without toxicity. Search for co-medication was negative. In 2019, the patient reported regular licorice ingestion. Two weeks after stopping this habit, ASOX levels were within the therapeutic range. Interestingly, this strong ABZ exposition for 2 years led to excellent control of AE lesions. Conclusion : clinicians in charge of AE patients have to be aware of these life habits that may impact on ABZ metabolism and consequently on disease evolution and/or drug toxicity.

1 : Pawluk S.A et al. Clin Pharmacokinet 2015;54:371-83.

#### IN VIVO EFFICACY OF ALBENDAZOLE FORMULATED AS SOLID DISPERSIONS AGAINST ECHINOCOCCUS MULTILOCULARIS

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The treatment of human alveolar echinococcosis consists of surgery and/or chemotherapy with benzimidazoles-methylcarbamates, mainly albendazole (ABZ). This drug has limited efficacy due to its poor and erratic bioavailability, attributed to its low dissolution rate. Solid dispersions (DSs) are considered a successful strategy to improve the dissolution profile and consequently the absorption of poorly soluble drugs such as ABZ. DSs are defined as the dispersion of one or more drugs in an inert carrier or matrix in the solid state. Recently, ABZ DSs have been developed using Poloxamer 407 (P407) as a carrier. This formulation significantly increased the solubility and dissolution rate of ABZ. The aim of the present work was to evaluate the clinical efficacy of ABZ:P407 DSs in mice infected with *Echinococcus multilocularis*. Fifty CF-1 mice were infected intraperitoneally with metacestodes of *E. multilocularis*. Six weeks post-infection, the mice were divided into 5 groups: 1- Water control; 2- P407 control; 3- ABZ-carboxymethylcellulose suspension; 4- Physical mixture (ABZ+P407) and 5- ABZ:P407 DSs. The treatments were performed orally, every 24 hours for 30 days. In all cases, the ABZ dose was 25 mg/kg. At the end of the treatment (approximately 10 weeks post-infection), mice were euthanized and the metacestodes were recovered. The efficacy of treatments was evaluated by comparing the weight of the cysts and by the ultrastructural

alterations studied by scanning electron microscopy. No significant differences were observed between the weight of the cysts recovered from the control groups ( $P>0.05$ ). All the treatments decreased the development of the cysts. However, only the mice treated with the ABZ:P407 DSs showed a significant decrease in the weight of the cysts ( $P<0.05$ ) compared to the control groups. These results coincide with the damage detected at the ultrastructural level. In conclusion, the formulation of ABZ as DSs increased the clinical efficacy of the drug.

## EFFECT OF PREOPERATIVE HBV-DNA LOAD ON SURGICAL TREATMENT OF HEPATIC ALVEOLAR ECHINOCOCCOSIS

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**Objective:** To study whether HBV-DNA load can influence the treatment effect and complications for patients with hepatic alveolar echinococcosis (AE) and chronic hepatitis B (HBV). **Material and Method:** The patients with AE and chronic hepatitis B in our center from January 2014 to July 2018 were enrolled as the test group. Twenty patients with AE only were treated with surgery as the control group. According to the HBV-DNA load, the test group were divided into group A, B and C, and the clinical characteristics and complications of different HBV-DNA load groups were compared. **Results:** Serum total bilirubin, prothrombin time, postoperation complications and hospitalization cost exhibited significant difference between test group and control group ( $P<0.05$ ). The infection of hepatitis B virus and the long diameter of the lesion is an independent risk factor for postoperative complications in patients with hepatic alveolar echinococcosis. There was no significant difference in postoperative liver function indexes and complications between the three groups ( $P>0.05$ ). **Conclusions:** 1. Hepatitis B virus infection has a certain impact on liver function indicators, hospital costs and complications of surgical treatment for patients with hepatic alveolar echinococcosis. 2. Preoperative infection with hepatitis B and lesion size were independent risk factors for postoperative complications.

**Key words:** alveolar echinococcosis; Hepatitis B virus; complication

## SURVEILLANCE OF HUMAN ALVEOLAR ECHINOCOCCOSIS IN FRANCE : DEVELOPMENT OF THE FRANCECHINO REGISTRY

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**Introduction.** The French National Reference Center for Alveolar Echinococcosis (CNR-EA), renamed National Reference Center for Echinococcosis (CNR-E) in 2017, has been providing national epidemiological surveillance for Alveolar Echinococcosis (AE) since 2012. We present the elements of this activity. **Material and Method.** The French AE Registry (FrancEchino) collects epidemiological and medical information on patients with AE in France since 1982. This collection makes it possible to study in particular the geographical distribution, the risk factors, the circumstances of diagnosis, the management and survival of patients. Since 2016, the information system has been available to a European network of partner centers (EurEchino Network) for the establishment of a European base (EurEchinodataBase), with a view to future expansion to an international registry on AE. **Results.** Among the new developments in the surveillance of AE, there is a steady increase in the number of cases recorded (17.2 incident cases / year in the period 2001-2006 vs 30.8 incident cases / year in the period 2006- 2012 vs 35.8 incident cases / year in the 2013-2018 period), an increase in cases diagnosed outside of the historical endemic area (25% of 2018 diagnoses vs 15% of 2017 diagnoses vs 9% in period 1982-2018), mainly in the North and West and recently in the South of France, and a very significant increase over the last 15 years in the number of AE diagnoses associated with an immunosuppression context (22% in the period 2004-2018 vs 5% before 2004). The circumstances of discovery also changed significantly, in favor of incidental diagnoses (55% in the period 2004-2018 vs 20% before 2004). **Conclusion.** The establishment and development of this surveillance system allow a dynamic assessment of the epidemiology, presentation and management of AE in France. Information on AE cases reporting, recording and data collection procedure is available on CNR-E website (<https://cnr-echinococcoses-ccoms.univ-fcomte.fr/>)

#### SIMPLE LIVER CYST OR CYSTOID LESION IN HEPATIC ALVEOLAR ECHINOCOCCOSIS: A RETROSPECTIVE COHORT STUDY WITH HOUNSFIELD ANALYSIS

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**Introduction.** Alveolar echinococcosis (AE) is a rare zoonosis caused by the larval stage of the fox tapeworm (*Echinococcus multilocularis*). AE affects the liver in about 98%. AE lesions have various morphological characteristics, described in the *Echinococcus multilocularis* Ulm classification for computed tomography (EMUC-CT). One of these

characteristics is a cystoid portion. Aim of the study was to compare the density of simple hepatic cysts with cystoid portions of AE lesions classified after EMUC-CT. Material and Methods. Hounsfield Unit (HU) density of cystoid areas of n=155 hepatic AE-lesions, classified after EMUC-CT was measured by two raters independently within the same CT-section. Measurements were performed in two different, most hypodense places, using a defined sized Region of Interest (ROI). The control group consisted of n=78 simple cysts. Results. HU measurement of cystoid portions of all EMUC-CT type I-IV lesions (n=155) gave a mean of 21.8±17.6, which was significantly different from that of 2.9±4.5 for the simple hepatic cysts (p<0.0001). The difference between each of the AE types and simple hepatic cysts was also significant. In addition, HU values of the cystoid portions in types I, II and IIIa/b and simple cysts were each significantly different from type IV (p<0.0001). HU measurements in type IV presented by far the highest mean. There was a high correlation between lesion size and density value. Discussion/Conclusion. The significantly higher density measured in the cystoid portions of hepatic AE lesions offers a good means of differentiation from simple hepatic cysts. The highest density of type IV, fits the structure of this lesion which, in addition to hypodense solid necrosis may also have a central alveolus varying in size as a sign of the vitality. Measurements may give hints for different stages of lesions.

Keywords: Alveolar echinococcosis, *Echinococcus multilocularis*, hepatic cysts, density, computed tomography, classification

#### HISTOLOGICAL ANALYSIS COMPARED TO COMPUTED TOMOGRAPHY MORPHOLOGIES IDENTIFIES DIFFERENT PATTERNS IN HEPATIC ALVEOLAR ECHINOCOCCOSIS POINTING TO A SEQUENTIAL EVOLUTION

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Introduction. Alveolar echinococcosis (AE) is caused by the larval stage of *Echinococcus multilocularis*. AE liver lesions are stratified into five different primary morphological types according to the *Echinococcus multilocularis* Ulm classification for computed tomography (EMUC-CT). The present study correlated CT data with histological parameters and aimed to identify histological characteristics of the various EMUC-CT types. Material and Methods. 45 samples of 44 patient were classified by means of the EMUC-CT. The accordingly classified cohort was analyzed regarding various histological parameters such as morphology of the central and peripheral part of the lesion comprising the laminated layer and the distribution of immunohistochemically detected small particles of *Echinococcus multilocularis* (spems) by the monoclonal antibody Em2G11. Results. The various EMUC-CT types revealed significant differences regarding the identified histological parameters. Type III lesions showed a pronounced fibrotic rim

in the periphery ( $212.92 \pm 113.66\mu\text{m}$ ), a shorter distance between laminated layer and peripheral zone ( $756.60 \pm 969.42\mu\text{m}$ ) and a higher number of spems in and outside the lesion. In contrast, type IV lesions are characterized by a reduced fibrotic peripheral zone ( $102.74 \pm 37.06\mu\text{m}$ ), a greater distance between the laminated layer and the border zone ( $2037.75 \pm 968.43\mu\text{m}$ ) and a single necrotic area with a central alveolar structure comprising the laminated layer. For type I and II lesions, these values lie between the values of type III and type IV lesions. Histological analysis of type V lesions show strong covering calcification. Discussion/Conclusion. The results point to various patterns of interactions between the host and the parasite leading to the different EMUC-CT types. We hypothesize that the various EMUC-CT types and their distinct histological patterns of the larval state of *Echinococcus multilocularis* in humans result from sequential time points during the infection reflecting different stages of the disease with type IV as an early lesion.

Keywords: Alveolar echinococcosis, Echinococcus multilocularis, computed tomography, classification, histology

#### HEPATIC ALVEOLAR ECHINOCOCCOSIS: CORRELATION BETWEEN COMPUTED TOMOGRAPHY MORPHOLOGY AND INFLAMMATORY ACTIVITY IN POSITRON EMISSION TOMOGRAPHY POINTS TO A POSSIBLE SEQUENTIAL EVOLUTION OF THE LESIONS

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Introduction: The aim was to evaluate quantified PET/CT activity as the expression of the inflammatory reaction of lesions in hepatic alveolar echinococcosis (AE) related to their primary morphology and calcification pattern. Material and Methods: 18-FDG PET/CT examinations from 51 treatment-naive patients with hepatic AE were classified according to the *Echinococcus multilocularis* Ulm classification for computed tomography (EMUC-CT), which defines a primary morphology of a lesion and a pattern of calcification. AE lesions were differentiated according to whether the surrounding PET activity was increased or not. Increased PET activity was quantified using the standardised uptake value (SUV). Results: *We detected a significantly lower PET activity of type IV lesions compared with the other lesion types and for the patterns of calcification the also type IV associated significantly lower PET activity in lesions without calcifications compared with lesions showing feathery calcifications, focal calcifications, or diffuse calcifications.* SUV according to primary morphological type gave statistically significant differences between type IV and type I ( $1.8 \pm 2.2$  versus  $6.5 \pm 3.8$ ;  $p=0.0009$ ), type IV and type II ( $1.8 \pm 2.2$

vs.  $6.1 \pm 3.1$ ;  $p=0.0019$ ), and type IV and type III ( $1.8 \pm 2.2$  vs.  $6.6 \pm 4.4$ ;  $p=0.0069$ ). Comparisons between type I, type II and type III showed no significant differences ( $p>0.05$ ). Regarding *the calcification patterns the highest SUVs obtained overall were in lesions with feathery calcifications*. There were also significant differences in SUV activity between Em2+ negative and Em2+ positive findings ( $2.11 \pm 2.26$  vs.  $6.93 \pm 3.56$ ;  $p<0.0001$ ). Discussion/Conclusion: *Type IV lesions appear to correspond to an early stage of the disease*. As types I, II and III show significantly higher SUVs than type IV, we can assume that these types are different stages of more advanced lesions, whose activity is located at the edges, allowing further extension. *The highest SUVs obtained overall were in lesions with feathery calcifications. Resulting therapeutic consequences should be subject of further studies*.

Keywords: Alveolar echinococcosis, Echinococcus multilocularis, computed tomography, classification, positron emission tomography

#### COULD METFORMIN ACT THROUGH THE LYSOSOMAL RAGULATOR COMPLEX TO COORDINATE TOR INHIBITION IN ECHINOCOCCUS SP.?

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Metformin (Met) is an anti-hyperglycemic agent, which also exerts antiproliferative effects. Recent data reveal that Met activates AMPK and inhibits TOR (the catalytic subunit of TORC1) by multiple, mutually nonexclusive mechanisms that do not necessarily depend on the inhibition of electron transport chain and the cellular ATP level. Among these, the assembly of the v-ATPase-Ragulator-AXIN/LKB1-AMPK complex on the lysosome surface appears to be of greatest significance at therapeutically achievable concentrations of this drug. We have previously shown that Met exhibits considerable in vitro and in vivo activity against E. granulosus larval stage. Here, we extended the study and evaluated the effect of the drug on E. multilocularis. Metformin partially arrested the in vitro differentiation of protoscolecocytes into metacestode vesicles and exerted a dose-dependent effect on the viability of parasite primary cells. At the molecular level, the drug induced depolarization of the mitochondrial membrane, activation of AMPK (increased Em-AMPK $\alpha$ -P176), suppression of Em-TOR (decreased Em-TOR-P3122) and overexpression of the autophagic marker Em-Atg8 in the germinal layer of in vitro cultivated metacestode vesicles. Besides, while insulin increased the levels of active Em-TOR, rapamycin reduced them in larval vesicles. Based on our in vitro results, we then examined the in vivo effect of Met on the growth of E. multilocularis larval stage in mice. Oral administration of Met (50 mg/kg/day for 8 weeks) was effective in achieving a significant reduction of parasite weight ( $1.5 \pm 1.1$  g) compared to untreated group ( $3.14 \pm 1.1$  g). Finally, by in silico assays, the presence of the key lysosomal pathway components were confirmed in the parasite. These results suggest that Met inhibits the development of Echinococcus spp. through AMPK activation and TOR inhibition and raise the question of whether this occurs indirectly, as a consequence of ATP synthesis inhibition, and/or directly, by the lysosomal pathway.

#### METABOLIC IMPACT OF HOST GLUCOSE HOMEOSTASIS CONTROLLING HORMONES ON THE ECHINOCOCCUS LARVAL STAGE

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Glucose availability is essential to ensure development processes in *Echinococcus granulosus*, both in definitive and intermediate hosts. Interestingly, the glucose modulating drugs niclosamide and metformin have proven anticestodal activity in human and mice, respectively. Both drugs, with an excellent safety profile in mammals, induce mitochondrial uncoupling in treated cells, and can improve insulin sensitivity and suppress glucagon signaling in treated hosts. In order to contribute to the understanding of in vivo anthelmintic mechanisms of metformin and the parasite-host hormonal interrelationship, we evaluated the metabolic activity of the parasite in presence of glucagon, insulin and somatostatin, host hormones for which the cestode expresses putative receptors. Four glucose transporters (GLUTs, capable of incorporating the 2-D-glucose fluorescent derivative) and two G-protein-coupled receptors with high sequence identity to human glucagon and somatostatin receptors were identified in the parasite larval stage. Although previous investigations have failed to demonstrate the gluconeogenesis in *Echinococcus* spp., we analyzed the transcriptional expression of glucose-6-phosphatase, fructose-1, 6-bisphosphatase, phosphoenolpyruvate carboxykinase and malate dehydrogenase in hormone-treated metacestodes and protoscoleces. In presence of exogenous insulin and glucagon, antagonistic effects in relation to glycogen content, free glucose levels and glycolytic flux were recorded in the parasite. While in response to glucagon, glycogenolysis and mitochondrial activation were induced in calcareous corpuscles, in presence of insulin, glycogen accumulation and AKT-TOR pathway activation were evidenced. This suggests that the parasite responds to host neuroenteropancreatic hormones. These peptide hormones may promote the parasite glucose homeostasis, necessarily required in the strobilar and vesicular development during the establishment of primary and secondary echinococcosis, respectively.

#### COMBINED TREATMENT WITH METFORMIN IMPROVES THE THERAPEUTIC EFFICACY OF LOW DOSES OF ALBENDAZOLE AGAINST EXPERIMENTAL ALVEOLAR ECHINOCOCCOSIS

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Chemotherapy treatment for echinococcosis has had limited effectiveness so far, therefore it is a dire need to find new drugs for its treatment. One of the most promising approaches to find new drugs against neglected diseases is the repurposing of compounds with already described activities against other pathogens. Metformin (Met) is an anti-hyperglycemic and anti-proliferative drug, which may exert a role in the therapy for multiple infectious diseases since it is effective on several pathogens. We have previously shown that the drug exhibits considerable in vivo activity against *E. multilocularis* larval stage. In mice intraperitoneally infected with 200 µl of metacestode material (8065 strain), the oral administration of Met (50 mg/kg/day) for 8 weeks from the day of infection was effective in achieving a significant reduction of parasite weight. Here, we extend the study and evaluate the in vivo efficacy of Met using different schemes of infection and treatment in the murine model of alveolar echinococcosis. First, we increased the parasitic inoculum to 500 µl of metacestode tissue. In this case, the oral administration of Met (100 mg/kg/day) combined with the lowest recommended dose of ABZ (5 mg/kg/day) for 8 weeks from the day of infection led to significant reduction in parasite weight compared with each drug alone. Subsequently, we started the treatments

5 weeks after the infection of mice with 200 µl of parasitic inoculum. Again, the combination of Met (100 mg/kg/day) together with low dose of ABZ (5 mg/kg/day) was highly effective in reducing the weight of parasite cysts compared with both drugs alone. In conclusion, based on our experimental data, Met emerges as a promising anti-echinococcal drug as it has proven to efficiently inhibit the development and growth of the *E. multilocularis* larval stage and its combination with ABZ may improve the current anti-parasitic therapy.

## 31 DE OCTUBRE

### IMMUNODIAGNOSIS OF CYSTIC ECHINOCOCCOSIS IN LIVESTOCK POPULATIONS: DEVELOPMENT AND VALIDATION OF AN ELISA KIT USING A RECOMBINANT B8/2 SUBUNIT OF ANTIGEN B OF ECHINOCOCCUS GRANULOSUS

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**Introduction:** The development of a high sensitivity and specificity screening tests for detecting cystic echinococcosis (CE) in livestock depends on the host-parasite interaction during the course of the infection, and the serological cross-reactivity with other taeniid cestodes. The taeniid 8-kDa protein family is comprised of 10 genes with high diversity that may provide evidence of cross-reaction or specific-reaction in the diagnosis of metacestode infection. We develop and validate an IgG ELISA kit using a recombinant B8/2 subunit of antigen B (rAgB8/2) of *Echinococcus granulosus*. Furthermore, the humoral response of sheep experimentally infected with 2000 *E. granulosus* eggs, and its evolution over time was evaluated during the first year. **Materials and Methods:** The rAgB8/2 was expressed as a fusion protein and purified by affinity chromatography. Evaluation of analytical and diagnostic performance of the method followed the OIE Manual procedures including serum panels of (n=500) naturally and (n=5) experimentally infected sheep; (n=50) animals immunized with EG95 vaccine, (n=900) uninfected lambs or (n=20) carrying other metacestode infections. **Results:** The ELISA kit using a rAgB8/2 was able to differentiate naturally and experimentally infected sheep population, from uninfected animals. The analytical and diagnostic performance of the method was validated successfully. The potential interference with vaccinated animals was removed and a specificity with < 1,8% cross reactivity was observed. The earliest detectable IgG response against rAgB8/2 was observed in sera from 2/5 and 4/5 sheep 7 and 14 days after experimental infection respectively. The highest immune response (5/5) have been found 16 to 24 weeks post infection and remained above the cut-off level until the end of the observation time. **Discussion:** The rAgB8/2-based ELISA could be recommended for screening of CE in sheep population specially in endemic areas. Coating with rAgB8/2 replaced successfully the use of hydatid fluid, infective material, and batch to batch variability.

**Key words:** Cystic echinococcosis, serodiagnosis, recombinant AgB8/2

THERAPEUTIC EFFECT OF IODINE 125 PARTICLE ON HEPATIC ALVEOLAR  
ECHINOCOCCOSIS IN THE ANIMAL MODEL  
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**Objective:** To investigate the therapeutic effect of iodine 125 (125I) particles in the animal model of alveolar echinococcosis (AE). **Methods:** Twenty nude mice were used to establish AE animal model and randomly divided into test group (n=10), control group (n=5) and blank control group (n=5). In the test group, the nude mice were implanted with 125I particle in the lesions; the control group underwent lesions puncture without particles implantation; the blank control group did not give any intervention. After 45 days of intervention, the lesion size and weighing were measured. The tissue samples were collected and examined by HE staining. The protoscolex were collected from the lesion and the activity was assessed by the trypan blue staining. **Results:** 1. The success rate of AE animal model in the nude mice was 80% (16/20). 2. After 45 days of 125I particles implantation, the diameters of the lesions among test groups, control group and blank control group exhibited statistically significant difference ( $P < 0.001$ ). 3. After intervention, the survival rate and activity of protoscolex in the test group was lower than the control group and the blank control group. **Conclusion:** 125I particles could kill protoscolex and inhibit the activity in AE animal model, and maybe used to treat AE.

IDENTIFICATION AND QUANTIFICATION OF PROTEINS INVOLVED IN CELL  
DIFFERENTIATION PROCESSES IN GERMINAL CELLS OF ECHINOCOCCUS  
GRANULOSUS

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Cystic echinococcosis is a severe neglected zoonotic infection caused by the larval stage (metacestode) of the parasite *Echinococcus granulosus* (Eg), which has a worldwide distribution and important medical and economic impact. The inner layer of the hydatid cyst or germinal layer contains a group of cells called germinative cells which are responsible for cell proliferation and from which differentiated cell types develop. Using a proteomic strategy, we identify and quantify proteins involved in processes of cellular differentiation in Eg germinative cells grown in different conditions. Eg primary cell culture was obtained from hydatid cysts and maintained with weekly splitting during 1-4 month. Cells were culture in normal media and conditioned media (resembling host environment) for different time periods. Then, 50  $\mu$ l of cells were homogenized in lysis buffer and the peptide content was estimated. Aliquots of 100  $\mu$ g protein per sample were used for filter-aided sample preparation and the resulting peptides were subjected to nano-LC-MS/MS-analysis. A combined library was set-up by combining the different runs using the Protein Pilot-software and Uniprot database. We identified a total of 455 proteins in all the conditions studied. We identified and quantified 9 proteins associated with cell differentiation which were differentially represented in the conditions studied. For example, HSP 90 alfa (Accession W6UM50) and Neurogenic notch (Accession W6UG53) were found overrepresented in cells cultured in normal media with a fold change 3,2 and 4,8 respectively ( $p < 0,05$ ) in comparison with cells culture in conditioned media (resembling host environment). Through the methodology used, it was possible to determine several proteins associated with cell differentiation present in the germinative

cells of Eg and to hypothesize its possible function. Moreover, many of the identified proteins could be useful as cestode developmental markers.

#### CHARACTERIZATION OF CYSTS DEVELOPMENT IN AN EXPERIMENTAL MURINE MODEL OF HEPATIC CYSTIC ECHINOCOCCOSIS

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Cystic echinococcosis (CE) is a zoonotic infection caused by the larval stage of *Echinococcus granulosus*. The disease is characterized by long term growth of cysts most commonly in the liver and lungs. Animal models play an important role in the study for novel drugs, surgical approaches, and vaccine development. We have recently established an experimental murine model of hepatic CE. The aim of the present work was to characterize the development of cysts over time in CF-1 mice infected with *E. granulosus* protoscoleces via the portal vein. Female mice (n=36) infected with 500 protoescoleces were allocated into four groups: A) 4 months post infection (p.i.); B) 5 months p.i.; C) 6 months p.i.; D) 7 months p.i. The cystic development was monitored by ultrasound, then mice were euthanized and samples of liver and lung were taken for histopathological examinations. An increase in the infection rate was observed over time, reaching 70% of infected mice at 7 months p.i. According to the ultrasonographic features, more than 90% of the cysts detected were active. Although there was an increase in the number and size of cysts developed in relation to the time of infection, no significant differences were found between the groups (P>0,05). The development of cysts in the liver did not show preference for any lobe. Interestingly, between 10-30% of the animals belonging to groups B, C and D presented cysts in the lung. The murine model of hepatic CE presents similar characteristics to the disease in humans: the route of infection, the development of cysts in the orthotopic and primary infection organ, the parasites can cross the liver filter and infect the lung and the histopathologic features of the cystic lesion. This model could be useful for the study of host-parasite interactions as well as drug efficacy trials.

#### BIOMOLECULAR ANALYSIS OF ECHINOCOCCUS GRANULOSUS CYSTS FROM SARDINIAN HUMAN PATIENT

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The purpose of our study was to characterize the larval form of *Echinococcus granulosus* by biomolecular analysis. Imagine-technique investigations were performed on 34 patients suspected to be affected by Cystic Echinococcosis (CE) from Sassari and Nuoro Hospital Wards. All sera were screened by “ELISA-Echinococcus IgG” (DRG-Germany)

and “Echinococcus-Western blot (WB) IgG” (LDBIO-France). Hydatids were excised from 7 patients (HU1 -> HU7). Cysts were inspected, then parasite material was observed by microscope, finally, analysed by biomolecular tests. PCR E.g.s.s. [primers for Calreticulin (Cal) gene-fragment of 1001bp (F5':CAATTTACGGTAAAGCAT-3' R5':CCTCATCTCCACTCTCT-3)], was performed on DNA extracted using DNeasy Blood and Tissue Kit (Qiagen, Germany). Afterwards, amplicons generated by PCR [primers for Cytochrome Oxidase 1 (COX1) fragment of 800bp (F5'-TTTTTTGGCCATCCTGAGGTTTAT-3' R5'-TAACGACATAACATAATGAAAATG-3')], were sequenced for genotype and haplotype determination. A Maximum Likelihood Phylogenetic Tree was built on a dataset comprising 4 human DNA isolates (HU1 -> HU4) and other reference sequences of *E. granulosus* s.l. retrieved from GenBank. Moreover, a Haplotype Network was calculated on the 4 human and the 83 DNA sequences of *E. granulosus* isolated from different animal species in Sardinia. Ultrasound techniques revealed one or more cysts attributable to CE on 23 patients, of whose 21 were localized in the liver and 2 in the lungs. According to WHO classification 2 (HU1, HU2) were active, 2 (HU3, HU4) transitional, and 3 (HU5 -> HU7) inactive. ELISA-DRG and WB-LDBIO evidenced 14 CE positive sera. The microscopy evidenced non-viable protoscolices in 4 cysts (HU1 -> HU4): 2 active and 2 transitional, of the 7 analysed. The remaining 3 presented necrotic materials. Biomolecular analysis identified *E. granulosus* s.s., G1 genotype for 2 cysts (HU2, HU4) and G3 genotype for the others 2 (HU1, HU3). Haplotype Network displayed evidence that 2 human DNA sequences were common to other animal species, while 2 were found only in human.

#### REGIONAL CONSTRUCTION OF MULTIMODAL IMAGING BIOBANK OF HYDATID DISEASE IN THE ERA OF PRECISION

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**Objective** Hydatid disease is a zoonosis seriously affecting the population in western China . Qinghai Province is a highly endemic area of hydatid disease . Relying on Qinghai University Affiliated Hospital , Association of Clinical Echinococcosis of Qinghai Province , Clinical Medical Research center For Hydatid Disease in Qinghai Province and Qinghai Province Hepatobiliary Pancreatic Surgery Center , a regional and standardized multimodal imaging biobank of hydatid disease was established in Qinghai Province . To explore the collection , preservation of multimodal imaging data, construction method , management and quality control of multimodal imaging biobank of echinococcosis . **Methods** Since 2011 , the patients' multimodal imaging including B-scan ultrasound , CEUS , CT ,MRI , 18F-FDG PET/CT(optional) and clinical informations including laboratory examination , therapeutic schedule , pathological imaging data and following-up data of hydatid disease were systemically collected and saved as a imaging biobank . At the same time, a random quality test is carried out on the sampled samples. **Results** Up to June 2019, a total of 1246 patients of hepatic cystic echinococcosis , 535 patients of hepatic alveolar echinococcosis and 268 patients of extrahepatic echinococcosis were collected . There are 18 cases with simultaneous hepatic alveolar and cystic echinococcosis , of which one case included situs inversus totalis . In addition, the quality detected by random sampling are in accordance with the quality standards of samples, which can meet the needs of scientific study. **Conclusion** We have preliminary established standardized biobank operation and management procedures , improved the efficiency of imaging data utilization, and provided multimodal imaging of data for basic and clinical research of hydatid disease .Multimodal imaging biobank is the important and effective

means of medical imaging data retrieval and analysis in clinical diagnosis, education and research.

Key words Hydatid disease ; Multimodal imaging ; Biobank ; Quality control ; Data management

## GENETIC CHARACTERIZATION OF HUMAN HYDATID CYSTS IN URUGUAY, SOUTH AMÉRICA

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**Introduction:** Cystic echinococcosis (CE) is a global health problem, considered a neglected disease by the World Health Organization. It is endemic and hyperendemic in most South America, especially Uruguay, Argentina, South of Brazil, Chile, Peru and Bolivia. CE is caused by the larval stage of *Echinococcus granulosus sensu lato* (*E. granulosus s. l.*). *E. granulosus s. l.* is composed for several species/ genotypes that differ in biological characteristics affecting the epidemiology and the control of this zoonosis. Among them, *E. granulosus sensu stricto* is composed by the G1 and G3 genotypes. The G1 genotype is the most frequently found worldwide and produces fertile hydatid cysts mainly in sheep but also in cattle and is frequently isolated from humans. The objective of this study was to identify the species/genotypes of *E. granulosus s. l.* that cause human CE in Uruguay. **Material and Methods:** A total of 13 samples of human hydatid cysts (HC) were analysed. Genomic DNA was obtained with commercial kit, following by Polymerase Chain reaction (PCR) and successive sequencing. **Results:** The results revealed the presence of *Echinococcus granulosus sensu stricto* in all cysts analyzed, which were identified as G1 genotype (N=12) and G3 (N=1). Two samples, identified as G1, were reported in children under 15 years of age. **Conclusions:** Despite the control program developed in Uruguay for decades, the results obtained could explain the persistence of the disease by the cycle dog- cattle, since cattle but no sheep are farmed in our country. Of note there is a particular active cycle of cases occurring in children under 15 years of age, indicating the persistence of the transmission in our country. Due to the diversity of species/ genotypes existing in the region it is relevant to continue with this kind of studies in order to obtain epidemiological conclusions.

## EMSB MICROSATELLITE GENOTYPING ON ISOLATED EGGS OF ECHINOCOCCUS MULTILOCULARIS : GENETIC DIVERSITY IN A DEFINITIVE HOST AND AT THE LOCAL LEVEL

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Abstract

**Introduction:** *Echinococcus multilocularis* (Em) is a cestode responsible for alveolar echinococcosis. The definitive host carnivores shelter adult worms that produce eggs

(infective forms of rodents and humans) which are spread in the environment with feces. The use of molecular markers makes it possible to identify and track parasitic genetic variants in the carnivore population. However, DNA is often extracted from adult worms involving trapping and autopsy. We developed a non-invasive approach that consists in typing one by one the eggs contained in feces of fox, dog and cat tested positive by an Em specific qPCR and to study their genetic variability using the microsatellite EmsB. Material and Method: Em eggs were concentrated by sucrose flotation and taken one by one with a micromanipulator (Leica microsystems) in carnivore feces (70 fox, 15 dog, 6 cat) collected in a village located in an endemic area of alveolar echinococcosis (Doubs department, France) and tested positive by qPCR. The EmsB profiles obtained from each egg were compared to a collection of European EmsB samples genotyped. Results: Em eggs were observed and isolated in 1/3 (31/96) carnivores feces, the majority of which are fox feces (N=30). In preliminary analyzes, 21 eggs isolated from 7 fox feces (1 to 4 eggs per feces) had 3 different EmsB profiles previously reported in the study area, in France and in Europe. Discussion/ Conclusion: These results indicate that the presence of Em DNA in faeces might not prove the presence of eggs. Therefore, confirmation by flotation is necessary to better estimate the risk of infection. Based on this non-invasive approach, the individual isolation of eggs made it possible to highlight EmsB profiles from feces for the first time.

Keywords: *Echinococcus multilocularis*, feces, EmsB

#### PROTEOMIC ANALYSIS OF ANTIGENIC PROTEINS OF *Echinococcus granulosus* AS VACCINE CANDIDATES

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Cystic echinococcosis is a zoonotic disease widely distributed throughout the world, which is caused by the larva of *Echinococcus granulosus*, affects livestock and humans. Canine is the definitive host, adult cestode resides in the small intestine, thus becoming the essential source for the occurrence of infection. Since the interruption of the infection cycle from the definitive host is required, it is necessary to study antigenic proteins in order to develop a potential vaccine. The aim of this research was to perform the proteomic analysis of antigenic proteins of excretory/secretory products obtained by in-vitro culture and hydrosoluble proteins of larval and adult stage of *E. granulosus*. The proteins were separated by two-dimensional electrophoresis, Western Blot (WB) was performed with serum and saliva from immunized dogs to identify antigenic proteins and these were sequenced by MALDI-TOF/TOF. A total of 12 spots were identified in this study as a result of the comparison of spot patterns on WB membranes of pre and post immunization serum and saliva. Sequence analysis revealed that 12 spots belonged to 5 known proteins of *E. granulosus* with structural, regulatory and enzymatic functions: Paramyosin, P29, Calumenin-B, Cytidine desaminase and protein 14-3-3. In the present study, antigenic proteins were identified using samples from previously immunized dogs, and these proteins were found in products obtained from both the larval and adult stages of the parasite. Using them as candidates for the design of a recombinant vaccine for the definitive host could affect the implantation, metabolism and development of the parasite, as well as help in the design of better diagnostic tests for the disease.

Key words: *Echinococcus granulosus*, excretory/secretory products, antigenic protein, MALDI-TOF/TOF

GENOTYPING OF *ECHINOCOCCUS GRANULOSUS* ISOLATES FROM CLINICAL SAMPLES OF PATIENTS ADMITTED TO EGE UNIVERSITY HOSPITAL BETWEEN 2016-2019

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**Introduction:** Human cystic echinococcus (CE) is a severe zoonotic disease caused by the larval stage of the tapeworm (cestode) *Echinococcus granulosus* whose strains (G1-G10) have highly importance due to difference in life cycles, host specificity, transmission dynamics etc. Although human CE is highly endemic in Turkey, there is lack of accurate data about genotypes of the *E. granulosus* complex that infect humans. The current study aimed to survey molecular characterization of *E. granulosus* isolates from human clinical samples by sequencing the mitochondrial genes of Cytochrome C Oxidase I (cox 1). **Material and Method:** The hydatid cyst materials of 23 patients admitted to Ege University Hospital and underwent surgery or Puncture, Aspiration, Injection, Re-aspiration (PAIR) procedure between November 2016- August 2019 were included in the study. Fertility of all cysts were confirmed by light microscopy. The total genomic DNA was extracted from protoscolexes by using Tissue-cell Genomic DNA Purification Kit according to manufacturer instructions with few modification. PCR was performed to amplify around 450 base pair (bp) for cox1 genes. PCR products were sequenced by Sanger sequencing and genotype diversity and sequence variation of the strains were analyzed via software named CLC Genomic Workbench 12.0.2.) to determine the genotypes. **Results:** The cox1 gene was successfully amplified in 20 out of 23 *E. granulosus* isolates. Sequencing of the amplified products revealed the presence of G1 as dominant genotype with 13 isolates, G2 and G3 in one isolate each. All of them were 99-100% identical to previously reported G1, G2 and G3 haplotypes. In addition, 4 isolates showed more than 99% similarity with both G1 and G2 and one isolate was 99.33 % identical with G1-G3 genotypes. **Conclusion:** The results showed that *E.granulosus sensu stricto* (G1-G3 complex) remains the most widespread genotype infecting humans in Turkey according to the some of the research results had been done until now. These finding might be useful for mapping of *E. granulosus* in revealing the regional transmission patterns and also in establishing control program. However, further molecular studies including larger sample sizes and other additional regions of Turkey are urgently required.

PURIFIED LAMINAR LAYER AND HYDATID FLUID OF ECHINOCOCCUS GRANULOSUS MODULATES DENDRITIC CELLS IMPROVING GLOBAL TRANSLATION BY ACTIVATING MTOR PATHWAYS AND PROMOTES CELL MATURATION AND T CELL PROLIFERATION.

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**Introduction:** The target of rapamycin (mTOR), represents a biological switch in the modulation of cell metabolism and environmental signals. It has recently been recognized as a regulator in the immune system. The aim of this work is to analyze if Echinococcus antigens could trigger an immune response by activating mTOR pathway in dendritic cells. **Methods:** BMDCs were cultured in complete RPMI. Hydatid fluid (HF)

was punctured from the hydatid cysts collected of infected cattle slaughtered. The germinal layer was removed from purified laminar layers (pLL) by washing with 2M NaCl. Cytotoxicity, phagocytosis, maturation of BMDCs and T cell proliferation were analyzed by FACS (mAbs directed to CD11c, CD40, CD80, CD86, MHC I and MHC II were used). Translation and phospho-mTOR were evaluated by Immunoblotting and confocal microscopy. Results: First, we evaluate cytotoxicity. Neither of the two antigens induce loss of cell viability. Protein synthesis was monitored using puromycin labeling. Cells not treated with puromycin, cycloheximide or rapamycin were used as controls. Both stimulus, pLL (MFI: 979) and HF (MFI:835), showed an increase in translation levels compared to its counterpart without stimulation (MFI:686, n=3). Furthermore, we have observed that DCs cultured without SFB or FLT3L, and stimulated with pLL or HF induce an increase in mTOR phosphorylation levels compared to untreated CDs (n=3, p=0,035 Anova test). Next, we evaluated the phenotype of DCs, both antigens downregulate phagocytosis levels (CTRL:80% vs PLL:75% or HF:66% n=3) and pLL of induce an up-regulation of MHCII and CD86 (n=3, p<0.05 vs Ctrl). There were no significant changes in CD40, CD80, and MHCI. Finally, we observed that culture T cells in the presence of stimulated DC induce their proliferation (%CFSE+ cells CTRL:99%, pLL:62%, HF:55% n=3). Conclusions: These data suggest that Echinococcus antigens induce an activation of mTOR pathway favoring global translation, their maturation and antigen presentation.

#### PROTEOMIC ANALYSIS OF EXOSOMES FROM PLASMA OF PATIENTS AFFECTED BY ACTIVE OR INACTIVE ECHINOCOCCUS GRANULOSUS CYSTS

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**INTRODUCTION:** Ultrasound is the reference diagnostic tool, weakly supported by serology when imaging is inconclusive. The availability of circulating biomarkers would considerably improve the diagnosis and much significantly the cyst staging (active vs inactive), treatments and follow-up. **MATERIALS and METHODS:** Plasma pools (~25ml) from patients (~25 subjects) infected either by active or inactive hydatid cysts, and two control pools were processed to isolate exosomes for proteomic label-free quantitative analysis. Results were statistically processed and clustered to group proteins related to either Active Cyst pool samples (APhs) or Inactive (IPhs) ones or shared by both samples or with controls. APhs and IPhs were subjected to bioinformatics analysis using STRING and Dynet Apps on Cytoscape platform. **RESULTS:** The graph shows great density and short radius. Several shared proteins at the centre confirmed the overlap of the majority of connected patterns, mostly associated to the Th1/Th2 response. However, several group-specific proteins showed high degree and centrality parameters, such as SRC, TGFB1 and some Integrins in APhs; and CDC-42, and Rab proteins in IPhs. GO Biological Process enriched STRING networks for immune effector response, wound healing, phagocytosis and, most evidently of all, the "Interspecies interaction between organisms" were centred on Cdc-42 in the case of IPhs and on Src tyrosine kinase in the case of APhs. **DISCUSSION/CONCLUSIONS:** Our results suggest the presence of different patterns elicited by *E. granulosus* to modulate the immune response and, in particular, Th1/Th2 balance, chemotaxis, cell polarity and migration involved in inflammatory pathways. In patients with active CE, a more finely tuned mechanism related to tyrosine kinases Src and Lyn and tyrosine phosphatase CD148 regulation seems to be in place. In

the presence of inactive CE, a more stable pattern which results directly in the Rac/Cdc42 activation and actin remodelling seems to be induced. This work was supported by the EU FP7, HERACLES project (GA602051).

COMPARISON OF FOUR COMMERCIAL KITS FOR THE DETECTION OF ANTIBODIES AGAINST ECHINOCOCCUS GRANULOSUS IN HUMAN SERA  
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Our purpose was to evaluate and compare 4 commercial kits for the detection of antibodies against Echinococcus granulosus antibodies in human sera: 1. "VIRapid-Hydatidosis" (Vircell, Spain), 2. "ELISA-Echinococcus IgG" (DRG, Germany), 3. "Echinococcus Western Blot (WB) IgG" (LDBIO-Diagnostics, France), 4. "CHORUS Echinococcus IgG" (Diesse, Italy). For the method performance Analytic Sensitivity (ASe), Reproducibility, Repeatability Accuracy and Precision were evaluated. For this purpose, firstly, 2 Cystic Echinococcosis (CE) positive sera were two fold serially diluted, their total protein quantified by Pierce™ BCA Protein-Assay-Kit (Thermo Scientific™), finally each dilution point was analyzed by each kit. Afterward, a total of 28 sera belonging respectively to 14 human patients affected by CE and 14 from free healthy donors were tested in duplicate analysis the 4 kits by two different operators. Evaluations were performed according to OIE and Italian-Accreditation-Body (ACCREDIA) procedure (Regulation UNI-CEI-EN-ISO/IEC 17025). Agreement between results was calculated by Cohen coefficient ( $\kappa$ ), according to Byrt table. ASe was similar for CHORUS-Diesse and ELISA-DRG and higher for WB-LDBIO and even more for Hydatidosis-VIRapid. The last test was still able to detect a weak signal at total protein concentration of 80 $\mu$ g/ml. Reproducibility performed by two operators, Repeatability, Accuracy and Precision evaluated by means of duplicate analysis of the 4 tests displayed a high agreement value of  $\kappa$ . Conversely, when results were compared to clinical data the agreement showed a fair  $\kappa$  value (0.427 to 0,620). Only 8 patients, out of the 14 CE clinically positive, resulted positive by all assays. Moreover, 1, having a surgically excised cyst, was negative to CHORUS-Diesse. Concerning WB-LDBIO, 1 patient, with a transitional hydatid, resulted negative and another 1, with a transitional was positive. Finally, 3 patients with, respectively, transitional, inactive and cerebral hydatids, were negative to all tests.

#### FOLLOW-UP OF PEDIATRIC CYSTIC ECHINOCOCCOSIS IN TUNISIA: VALUE OF 2B2, 2B2T AND P29 RECOMBINANT ANTIGENS

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**Introduction.** Cystic echinococcosis (CE) remains a serious public health burden worldwide. The available serological immunoassays are not fully satisfactory for the follow-up of CE cases. The performance of 2B2, 2B2t and P29 recombinant (rec) antigens have been previously assessed on an adult patient cohort. The aim of this study was to evaluate the usefulness of these antigens in the follow-up of CE in Tunisian pediatric population. **Methods.** A total of 62 children operated for CE were followed for 1 year post-surgery. Based on the postoperative outcomes, patients were classified into: "Cured CE" (CCE) and "non-cured CE" (NCCE). Sera samples were tested by ELISA against the three rec proteins. We used the Wilcoxon test to compare the antibody levels over the follow-up time points. **Results.** Our results demonstrate that antibody levels decrease significantly one year post-surgery compared to levels measured in 1 month after surgical treatment. For the rec 2B2, 2B2t and P29, p-values were 0.0001, 0.008 and 0.003 respectively. **Controversy,** NCCE exhibit permanent high immunoreactivities during all the follow-up (all p value > 0.1). **Conclusion.** The decrease of antibody levels against rec P29, 2B2 and 2B2t one year after surgery in CCE and their persistence however in NCCE indicates that these proteins may be useful for the follow-up of CE cases.

**Key words:** Follow-up, children, Cystic Echinococcosis

#### "THORACOSCOPIC APPROACH OF PEDIATRIC PULMONARY HYDATIDOSIS' TREATMENT"

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**Introduction:** Hydatidosis is a pathology caused by *Echinococcus granulosus* sl., parasitic zoonosis endemic in Patagonia Argentina, in which man is intermediate host, hosting the larva or hydatid cyst (QH). These present different anatomical locations, being the lungs, the ones that represent 21% of the surgical cases registered in Comodoro Rivadavia Hospital, statistics department. In children, the clinical manifestations of pulmonary hydatidosis are earlier due to the lower lung capacity and / or the lower physiological reserve of the lungs compared to the adult. Treatment is normally surgical and involves enucleation and cystic excision through indirect marsupialization, a technique used by our team. **Objective:** To replicate the principles of conventional surgery by using videothoracoscopy as a minimally invasive technique. **Material and Method:** 5 patients were treated during a period of 36 months, average age 6 years, with QH of anatomical location in both lower lobes, four right and one left. Selective bronchial intubation causing atelectasis of the affected lung without producing pneumothorax. Due to the basal location, the first 10 mm trocar located sub axillary midline of the third intercostal space (EI) medium line CO2 pressure 3 to 4 mm Hg, the second 5 mm trocar level 4 EI The anterior axillary line and third trocar of 5 mm placed at the level of 5 EI posterior axillary line. Pulmonary release and cystic exposure, placement of soaked gauze with 0.9% physiological solution surrounding the cyst as prophylaxis to avoid accidental loss of hydatid fluid. Then with a laparoscopic puncture needle, the cyst is penetrated by aspirating its contents, having a second system in case of any unforeseen situation, later, aspiration of the hydatid membrane, isolating and placement of it in a polyethylene bag is needed, extracting it through port one. Vascular sealer with bipolar energy was used for both pleuroparietal adhesions and remains of the cyst. Hydrostatic test with

bronchorrhaphy if there is an eventual air leak. Results: thoracoscopy for lung cysts in children is feasible, and safe, reduces surgical aggression and offers better results. The described technique reduced the surgical average time to 73 min, hospitalization time of 5 days ( $\pm$  1), extraction of the thoracic tube between 2 and 4 days. No recurrence so far and no adverse side effects. In one of the cases, conversion to wide vertical axillary thoracotomy was required due to the large cystic size. Conclusion: videothoracoscopy is a reproducible and executable technique for the hydatid pulmonary cyst treatment in pediatric patients, causing less pain after the operation and getting better aesthetic results than conventional thoracotomy. The results obtained allow us to conclude that videothoracoscopy fulfills the objectives of the hydatidosis treatment offering additional benefits.

Key Words: Pulmonary Hidatidosis-Children-Videothoracoscopy.

CYSTIC ECHINOCOCCOSIS OF LOCAL ORIGIN IN LAMBAYEQUE OPERATED IN A  
PEDIATRIC HOSPITAL OF LIMA PERU, 2011-2015

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**INTRODUCTION.** Human cystic echinococcosis is an endemic parasitic zoonosis in Peru, caused by cestodes of the gender *Echinococcus granulosus*. The person acts like accidental intermediate guest, which is infected by direct contact with the fecal matter of parasitized dogs; or when consuming water, vegetables or fruits with eggs of the parasite. **OBJECTIVE.** Demonstrate cases of cystic echinococcosis of local origin in the Lambayeque Region. **MATERIAL AND METHOD.** Descriptive, retrospective and observational study. The study population consisted of 194 patients diagnosed with cystic echinococcosis operated at the National Institute of Child Health, 2011-2015, of which 2% came from the Lambayeque region. Data were obtained from the review of medical records and information provided by family members of patients. **RESULTS.** All cases register birth in the Lambayeque Region, and do not register travel to other regions of the country. The ages fluctuate between 6 and 12 years, male sex. The surgical techniques used were craniotomy (33%) and thoracotomy (67%). Histopathological results correspond to the presence of viable hydatid cyst membrane. The first case corresponds to cerebral cystic echinococcosis of Morrope (Lambayeque); hospitalized with diagnosis of craniopharyngioma. He was discharged with DAV sequels. They raise sheep and feed dogs with raw viscera. The second case corresponds to bilateral cystic echinococcosis, from Pomalca (Chiclayo); hospitalized with a diagnosis of lung tumor. They raise sheep and dogs. The third case corresponds to bilateral cystic echinococcosis, from Pimentel (Chiclayo); he is admitted with a diagnosis of bronchogenic cyst, with a history of residence in the district of José Leonardo Ortiz (Chiclayo). **DISCUSSION AND CONCLUSION.** Autochthonous cases of cystic echinococcosis in the Lambayeque region are demonstrated in places where the informal benefit and practices of feeding raw viscera of cattle to dogs are associated.

Key words: echinococcosis, surgical treatment, hospital pediatric.

THE STATE OF EUROPEAN REGISTER OF CYSTIC ECHINOCOCCOSIS (ERCE) 5 YEARS  
AFTER ITS INCEPTION

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Kamena VUTOVA, Mar SILES-LUCAS, Enrico BRUNETTI, Adriano CASULLI, on behalf of



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the ERCE network\*\*In alphabetical order: Andrea ANGHEBEN, Nikolay BAGMET, Moncef BELHASSEN, Solange BRESSON-HADNI, Fabrizio BRUSCHI, Guido CALLERI, Barbara CASTIGLIONI, Cathy CHEMLA, Leonardo CHIANURA, Balazs DEZSENYI, Maria Teresa GIORDANI, Valbona GJONI, Levan GOGICHAISHVILI, Delia GOLETTI, Majid HARANDI, Gulziya ISMAILOVA, Fazal KARIM, Emma LAPINI, Felix LÖTSCH, Scilla MASTRANDREA, Guido MENOZZI,, Andrè PAUGAM, Alfonso RECORDARE, Malgorzata SULIMA, Antonella TEGGI, Carlo TORTI, Giustina VITALE, Martine WALLON, Lorenzo ZAMMARCHI Patrizia ROSSI: Department of Infectious Diseases, Istituto Superiore di Sanità, Rome, Italy.

**INTRODUCTION:** Cystic echinococcosis (CE) is a neglected zoonosis and the real burden of infection is difficult to evaluate. Given the clinical features of human CE, only a fraction of clinically relevant cases reach medical attention, they are rarely notified and this lack of data contributes to the poor evidence base, which hampers evidence based guidelines for its management. The European Register of CE (ERCE), launched in October 2014 to address the problem is a prospective, observational, multicentre register of patients with CE. Its aims are to gauge the burden of human CE in Europe, bring CE to the attention of health authorities, encourage control policies, and support research on CE. We outline the state and perspectives of ERCE, 5 years after its inception. **MATERIALS AND METHODS:** The ERCE database was searched (31/03/2019) and data concerning patients' registration, follow-up visits, CE cyst details, and clinical management were analysed. **RESULTS:** 44 centres in 15 countries contribute to ERCE. Of these, 33 (75%) registered patients and, of these, 18 (54%) recorded at least one visit within the past 18 months. 2,043 patients were registered. 25 (51%) centres recorded follow-up visits after the first patient registration, with a median 25% registered patients having follow-up visits recorded. Cysts characteristics (organ/s involved and/or cyst stage for at least one patient/visit) were recorded by 28 centres. Clinical management details could be analysed for 726 cysts (in 523 patients), for 895 stage-management matched observations. **DISCUSSION/CONCLUSIONS:** ERCE has been constantly expanding, with several Centres contributing data. ERCE has successfully achieved the objective of highlighting the burden of CE in Europe, but number and quality of data need to be improved if all its ambitious goals are to be met.

**Keywords:** cystic echinococcosis, prospective register of cases, epidemiological and clinical surveillance

#### THE INTERNATIONAL IMPACT OF HERACLES COLLABORATIVE PROJECT ON CYSTIC ECHINOCOCCOSIS

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**INTRODUCTION:** Cystic Echinococcosis (CE) is one of the most important zoonotic diseases worldwide and was recently assigned to the list of the Neglected Tropical Diseases prioritized by the WHO. Tools for its diagnosis and treatment are currently not standardized, partly due to the complex and chronic evolution of CE and lack of funding to support prospective multicenter clinical trials, which in turn make data on this infection poorly framed and evidence supported, resulting in yet more neglect. **MATERIALS and METHODS:** The main goals were to: Identify the population affected by CE in Bulgaria, Romania and Turkey; create the European Register of CE (ERCE); establish a bio-sample repository; set-up and validate new recombinant antigens;

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identify cyst stage-specific biomarkers; increase drug bioavailability of benzimidazoles. RESULTS: Core achievements are: 1) Creation of the “Heracles Extended Network” with more than 60 centers from 30 countries ([http://www.heracles-fp7.eu/interactive\\_map.html](http://www.heracles-fp7.eu/interactive_map.html)); 2) Biggest research-based cross-sectional ultrasound-based population study (n=24,693) on CE, estimating 151,000 people infected in rural Romania, Bulgaria and Turkey; 3) Creation of the European Register as a prospective case series with more than 2,000 patients registered from 48 centres (<http://www.heracles-fp7.eu/erce.html>); 4) Patent on anti-parasitic soluble drugs: “Salts of compounds having a benzimidazolic structure” (PCT/IT2016/000191); 5) Creation of the Echino-Biobank to sustain experimental and clinical research in this field (n=5,000 samples used); 6) Worldwide collection of human cyst samples for genotyping studies (n=742); 7) First proteomic description of parasite exosomes in echinococcal cyst and identification of biomarker candidates in plasma by quantitative proteomic analysis; 8) Scientific papers published in peer-reviewed journals: 58. DISCUSSION/CONCLUSIONS: The results from HERACLES project (2013-2018) will support governments, international agencies, the European Commission, to harmonize data collection, monitoring and reporting of CE. We see this as breakthrough in the current scenario of CE. This work was supported by the EU FP7, HERACLES project (GA602051; <http://www.Heracles-fp7.eu/>).