



# TRYLEIDIAG PRESS REVIEW

## July 2008

### Content

#### Events Highlights and Grants

- *Fourth World Congress on Leishmaniasis Worldleish4 (2009) Central Drug Research Institute, Lucknow, INDIA*
- *Congress MEEGID IX in University of California, 30th October-1st November 2008 in Irvine, USA*

#### Research news

- *Prevalence of bovine trypanosomiasis in Central Mozambique from 2002 to 2005. Genetic analysis of the human infective trypanosome, Trypanosoma brucei gambiense: chromosomal segregation, crossing over and the construction of a genetic map.*
- *Trypanosomiasis-induced B cell apoptosis results in loss of protective anti-parasite antibody responses and abolishment of vaccine-induced memory responses.*
- *Paratransgenesis applied for control of tsetse transmitted sleeping sickness.*
- *Congenital Chagas disease: Diagnostic and clinical aspects in an area without vectorial transmission, Bermejo, Bolivia.*
- *Miltefosine for visceral leishmaniasis relapse treatment and secondary prophylaxis in HIV-infected patients.*
- *Leishmania donovani leishmaniasis in Cyprus.*
- *Tamoxifen Is Effective in the Treatment of Leishmania amazonensis Infections in Mice.*
- *Characterization of widespread canine leishmaniasis among wild carnivores from Spain.*
- *Clinical manifestations of cutaneous leishmaniasis in Sri Lanka - possible evidence for genetic susceptibility among the Sinhalese.*
- *Identification of geographically distributed sub-populations of Leishmania (Leishmania) major by microsatellite analysis.*
- *Inhibition by Dications of In vitro growth of Leishmania major and Leishmania Tropica: causative agents of old world cutaneous Leishmaniasis.*
- *Neglected infections of poverty in the United States of america.*
- *Leishmania infantum: tuning digitonin fractionation for comparative proteomic of the mitochondrial protein content.*
- *Macrophage and T-Cell Gene Expression in a Model of Early Infection with the Protozoan Leishmania chagasi.*
- *Accelerated Control of Visceral Leishmania donovani Infection in IL-6-Deficient Mice.*
- *Leishmaniasis of the tongue due to leishmania infantum.*
- *Imported tropical infectious ulcers in travelers.*
- *Cutaneous leishmaniasis of the lower lip in a United States soldier.*
- *Validation of a recombinant based antibody ELISA for diagnosis of human and canine leishmaniasis.*

#### Political and regulatory

- *"The right to Health" is published in an effort to explore the links between health care and human rights (published by Office of the United Nations High Commissioner for Human Rights (OHCHR), in collaboration with the World Health Organization)*
- *FIND celebrates five years of its successful program to combat neglected diseases with more efficient diagnostic tests*



---

## EVENTS HIGHLIGHTS

### **Fourth World Congress on Leishmaniasis WorldLeish4**

*February 3rd-7th 2009*

*Lucknow, INDIA*

In WorldLeish 4, the organizing committee hopes to re-unite leishmaniacs by providing a forum to all for presenting their state-of-the-art research with special emphasis on "Translational Health Science". Scientists from all over the world will have the opportunity to share their findings, thoughts, and progress. It is also necessary to provide a fresh impetus to the long-term aims to identify areas of interest for future studies, and to promote interactions and cooperation among scientists from all disciplines of relevance in order to solve the global problems of leishmaniasis.

More information: <http://www.worldleish4.org/>

### **Congress MEEGID IX**

*October 30th -November 1st 2008*

*Irvine, USA*

Topics: genetics, genomics, proteomics, population biology, mathematical modeling, and bioinformatics. Papers can report on the host, the pathogen (viruses, parasitic protozoa, helminths, fungal organisms, and prions), or the vector for vector-borne diseases. Papers considering co-evolution phenomena are particularly encouraged. All infectious models can be considered, including those of veterinary or agronomical relevance. Special emphasis will be given to health problems of particular interest to Africa and the Indian Ocean: AIDS, malaria, tuberculosis, sleeping sickness, ebola, chikungunya, cattle and crop pathogens.

More information: <http://www.th.ird.fr/meegid/meegid.htm>



## RESEARCH NEWS

### Prevalence of bovine trypanosomiasis in Central Mozambique from 2002 to 2005.

Specht EJ.

Laboratório Regional de Veterinária, P.O. Box 42, Chimoio, Mozambique. [vetlabman@teledata.mz](mailto:vetlabman@teledata.mz)

Onderstepoort J Vet Res. 2008 Mar;75(1):73-81.

The study is the result of analyzing 16895 blood smears of cattle collected at 180 sites in the provinces of Manica, Sofala, Zambézia and Tete in Mozambique. Of the blood smears 73.9% were from Manica, 11.8% from Tete, 8.5% from Sofala and 5.8% from Zambézia; 75.6% of these were collected from smallholder cattle. Infections with trypanosomes were highest in smallholder cattle from Sofala Province with 36.8% of the 872 blood smears examined positive for trypanosomes, and lowest in cattle of commercial farmers in Manica Province with only 6.2% of 2252 blood smears being positive. *Trypanosoma congolense* was the predominant species, followed by *Trypanosoma vivax* and *Trypanosoma brucei sensu lato*. *Trypanosoma brucei*, which also infects humans, was more frequent in the districts of Buzi, Mutarara and Morrumbala with 15.1%, 10.5% and 9.8% of all examined cattle in 2005 being infected with it, respectively. The results show a significant increase in the infection rate with trypanosomes compared with results obtained in previous years by the Regional Veterinary Laboratory in Manica Province and by the Regional Tsetse and Trypanosomiasis Control Programme in Zambézia, Tete and Sofala provinces.

### Genetic analysis of the human infective trypanosome, *Trypanosoma brucei gambiense*: chromosomal segregation, crossing over and the construction of a genetic map.

Cooper A, Tait A, Sweeney L, Tweedie A, Morrison L, Turner CM, Macleod A.

Genome Biol. 2008 Jun 22;9(6):R103. [Epub ahead of print]

**BACKGROUND:** *Trypanosoma brucei* is the causative agent of human sleeping sickness and animal trypanosomiasis in sub-Saharan Africa and has been subdivided into three subspecies: *Trypanosoma brucei gambiense* and *Trypanosoma brucei rhodesiense*, which cause sleeping sickness in humans and the non-human infective *Trypanosoma brucei brucei*. *T.b.gambiense* is the most clinically relevant sub-species, responsible for over 90% of all human disease. The genome sequence is now available and a Mendelian genetic system has been demonstrated in *T.brucei*, facilitating genetic analysis in this diploid protozoan parasite. As an essential step towards identifying loci that determine important traits in the human-infective subspecies, we report the construction of a high-resolution genetic map of the STIB 386 strain of *T.b.gambiense*. **RESULTS:** The genetic map was determined using 119 microsatellite markers assigned to the 11 megabase chromosomes. The total genetic map length of the linkage groups was 733.1 centiMorgans covering a physical distance of 17.9 Megabases with an average map unit size of 24 Kilobases per centiMorgan. Forty-seven markers in this map were also used in a genetic map of the non-human infective *T.b.brucei* subspecies permitting comparison of the two maps and showing that synteny is conserved between the two subspecies. **CONCLUSIONS:** The



genetic linkage map presented here is the first available for the human-infective trypanosome, *T.b.gambiense*. In combination with the genome sequence, this opens up the possibility of using genetic analysis to identify the loci responsible for *T.b.gambiense* specific traits such as human infectivity as well as comparative studies of parasite field populations.

## **Trypanosomiasis-induced B cell apoptosis results in loss of protective anti-parasite antibody responses and abolishment of vaccine-induced memory responses.**

Radwanska M, Guirnalda P, De Trez C, Ryffel B, Black S, Magez S.

*Laboratoire de Parasitologie, Université Libre de Bruxelles, ULB, Brussels, Belgium.*

PLoS Pathog. 2008 May 30;4(5):e1000078.

African trypanosomes of the *Trypanosoma brucei* species are extra-cellular parasites that cause human African trypanosomiasis (HAT) as well as infections in game animals and livestock. Trypanosomes are known to evade the immune response of their mammalian host by continuous antigenic variation of their surface coat. Here, we aim to demonstrate that in addition, trypanosomes (i) cause the loss of various B cell populations, (ii) disable the hosts' capacity to raise a long-lasting specific protective anti-parasite antibody response, and (iii) abrogate vaccine-induced protective response to a non-related human pathogen such as *Bordetella pertussis*. Using a mouse model for *T. brucei*, various B cell populations were analyzed by FACS at different time points of infection. The results show that during early onset of a *T. brucei* infection, spleen remodeling results in the rapid loss of the IgM(+) marginal zone (IgM(+)MZ) B cell population characterized as B220(+)IgM(High)IgD(Int)CD21(High)CD23(Low)CD1d(+)CD138(-). These cells, when isolated during the first peak of infection, stained positive for Annexin V and had increased caspase-3 enzyme activity. Elevated caspase-3 mRNA levels coincided with decreased mRNA levels of the anti-apoptotic Bcl-2 protein and BAFF receptor (BAFF-R), indicating the onset of apoptosis. Moreover, affected B cells became unresponsive to stimulation by BCR cross-linking with anti-IgM Fab fragments. In vivo, infection-induced loss of IgM(+) B cells coincided with the disappearance of protective variant-specific T-independent IgM responses, rendering the host rapidly susceptible to re-challenge with previously encountered parasites. Finally, using the well-established human diphtheria, tetanus, and *B. pertussis* (DTPa) vaccination model in mice, we show that *T. brucei* infections abrogate vaccine-induced protective responses to a non-related pathogen such as *B. pertussis*. Infections with *T. brucei* parasites result in the rapid loss of T-cell independent IgM(+)MZ B cells that are normally functioning as the primary immune barrier against blood-borne pathogens. In addition, ongoing trypanosome infections results in the rapid loss of B cell responsiveness and prevent the induction of protective memory responses. Finally, trypanosome infections disable the host's capacity to recall vaccine-induced memory responses against non-related pathogens. In particular, these last results call for detailed studies of the effect of HAT on memory recall responses in humans, prior to the planning of any mass vaccination campaign in HAT endemic areas.

## **Paratransgenesis applied for control of tsetse transmitted sleeping sickness.**

Aksoy S, Weiss B, Attardo G.

*Yale University School of Medicine, Department of Epidemiology and Public Health, New Haven, CT 06520, USA. serap.aksoy@yale.edu*



Adv Exp Med Biol. 2008;627:35-48.

African trypanosomiasis (sleeping sickness) is a major cause of morbidity and mortality in Sub-Saharan Africa for human and animal health. In the absence of effective vaccines and efficacious drugs, vector control is an alternative intervention tool to break the disease cycle. This chapter describes the vectorial and symbiotic biology of tsetse with emphasis on the current knowledge on tsetse symbiotic genomics and functional biology, and tsetse's trypanosome transmission capability. The ability to culture one of tsetse's commensal symbiotic microbes, *Sodalis* in vitro has allowed for the development of a genetic transformation system for this organism. Tsetse can be repopulated with the modified *Sodalis* symbiont, which can express foreign gene products (an approach we refer to as paratransgenic expression system). Expanding knowledge on tsetse immunity effectors, on genomics of tsetse symbionts and on tsetse's parasite transmission biology stands to enhance the development and potential application of paratransgenesis as a new vector-control strategy. We describe the hallmarks of the paratransgenic transformation technology where the modified symbionts expressing trypanocidal compounds can be used to manipulate host functions and lead to the control of trypanosomiasis by blocking trypanosome transmission in the tsetse vector.

## **Congenital Chagas disease: Diagnostic and clinical aspects in an area without vectorial transmission, Bermejo, Bolivia.**

Brutus L, Schneider D, Postigo J, Romero M, Santalla J, Chippaux JP.

*IRD UR010, Research Unit "Mother's and Child's Health in Tropical Environment", Institut de Recherche pour le Développement, La Paz, Bolivia.*

Acta Trop. 2008 Jun;106(3):195-9. Epub 2008 Mar 27.

The authors carried out a 1-year study of a population of pregnant women delivering at Bermejo hospital, South Bolivia. In this area, vectorial transmission of *Trypanosoma cruzi* is negligible and women infect themselves during displacements in close endemic areas. The prevalence of *T. cruzi* in 508 pregnant women, diagnosed by several serological tests, was 33.9%. In eight infants, we observed *T. cruzi* in the umbilical cord (congenital transmission rate of 5.2%). The means of birth weights, lengths and hemoglobin rates were similar in the children from both seronegative and seropositive women, and in children infected or not by *T. cruzi*. This study could confirm a less severity of the congenital disease of Chagas in the absence of re-infestation of the mother during pregnancy. Serological screening of pregnant women by rapid diagnostic tests and examination of babies born from seropositive mothers by microhematocrit method at birth is a suitable strategy to detect and prevent congenital Chagas disease in non-endemic areas.

## **Miltefosine for visceral leishmaniasis relapse treatment and secondary prophylaxis in HIV-infected patients.**

Marques N, Sa R, Coelho F, Oliveira J, Saraiva Da Cunha J, Melico-Silvestre A.

*From the Infectious Diseases Department, University Hospital of Coimbra, Coimbra, Portugal.*

Scand J Infect Dis. 2008;40(6):523-6.

S

Miltefosine is the first effective oral drug against visceral leishmaniasis. However, there are few data



about its role against the increasing problem of HIV-associated visceral leishmaniasis. It is necessary to establish a treatment and secondary prophylaxis approach with miltefosine in this population, particularly for those in whom standard treatment was unsuccessful. We report our experience with miltefosine in 5 HIV-infected patients. Miltefosine was used in relapse treatments (50 mg, b.i.d.) in 3 patients and as maintenance therapy (50 mg, 3 times/week) in all of them. Miltefosine was discontinued after full recovery of immune function in 4 patients. The median disease-free period has been 20 months since miltefosine discontinuation. One patient was lost to follow-up. Miltefosine dosage regimens for the treatment of relapses and for maintenance treatment in HIV-infected patients should be established in prospective studies.

## **Leishmania donovani leishmaniasis in Cyprus.**

Léger N, Depaquit J.

*USC VECPAR-AFSSA, Faculté de Pharmacie de Reims, Université de Reims Champagne-Ardenne, Reims, France.*

Lancet Infect Dis. 2008 Jul;8(7):402.

## **Tamoxifen Is Effective in the Treatment of *Leishmania amazonensis* Infections in Mice**

Danilo C. Miguel, Jenicer K. U. Yokoyama-Yasunaka, Silvia R. B. Uliana<sup>\*</sup>

*Departamento de Parasitologia, Instituto de Ciências Biomédicas, Universidade de São Paulo, São Paulo, Brazil*

PLOS Neglected Tropical Diseases, June 2008, Vol 2, 249

Chemotherapy is still a critical issue in the management of leishmaniasis. Until recently, pentavalent antimonials, amphotericin B or pentamidine compounded the classical arsenal of treatment. All these drugs are toxic and have to be administered by the parenteral route. Tamoxifen has been used as an antiestrogen in the treatment and prevention of breast cancer for many years. Its safety and pharmacological profiles are well established in humans. We have shown that tamoxifen is active as an antileishmanial compound *in vitro*, and in this paper we analyzed the efficacy of tamoxifen for the treatment of mice infected with *Leishmania amazonensis*, an etiological agent of localized cutaneous leishmaniasis and the main cause of diffuse cutaneous leishmaniasis in South America.

## **Characterization of widespread canine leishmaniasis among wild carnivores from Spain.**

Sobrino R, Ferroglio E, Oleaga A, Romano A, Millan J, Revilla M, Arnal MC, Triscioglio A, Gortázar C.

*Instituto de Investigación en Recursos Cinegéticos IREC (CSIC-UCLM-JCCM), Ronda de Toledo s/n, 13071 Ciudad Real, Spain.*

Vet Parasitol. 2008 May 10.



Visceral Leishmaniasis (VL) is an emerging zoonotic parasitic disease caused by *Leishmania infantum* in Mediterranean countries, with sand flies (*Phlebotomus* spp.) as vectors and dogs as the main domestic reservoir. The role of wild carnivores in the epidemiology of leishmaniasis is still controversial. In order to determine the prevalence of natural infection with *L. infantum* in wild carnivores from Spain, we analyzed 217 samples by PCR and western blotting and used restriction fragment length polymorphism (RFLP) to compare the patterns present in wild carnivores with those of domestic dogs from the same areas. DNA of the parasite was detected in spleen or blood samples from 35 (16.12%) analyzed wild carnivores, including 8 of 39 (20.5%) wolves (*Canis lupus*), 23 of 162 (14.1%) foxes (*Vulpes vulpes*), 2 of 7 (28.6%) Egyptian mongooses (*Herpestes ichneumon*), 1 of 4 genets (*Geneta geneta*), and 1 of 4 Iberian lynxes (*Lynx pardinus*). No significant sex or age differences in prevalence were observed in wolves and foxes ( $P > 0.05$ ), but there was a significant difference among regions in foxes ( $P < 0.05$ ). A total of 12 PCR-RFLP patterns were found in foxes, 6 in wolves, 4 in dogs, 2 in Egyptian mongooses and 1 in lynx and genet. RFLP patterns differed between dogs and foxes in the two areas where they could be compared. This is the first study of canine leishmaniasis in wild canids and other carnivores from different regions of Spain by PCR. The prevalence of infection indicates the existence of natural infection in apparently healthy wild carnivore populations, and our results are suggestive of a sylvatic cycle independent of dogs.

## **Clinical manifestations of cutaneous leishmaniasis in Sri Lanka - possible evidence for genetic susceptibility among the Sinhalese.**

Samaranayake TN, Dissanayake VH, Fernando SD.

*Department of Parasitology, Faculty of Medicine, University of Colombo, 271 Kynsey Road, Colombo 8, Sri Lanka.*

Ann Trop Med Parasitol. 2008 Jul;102(5):383-90.

Human cutaneous leishmaniasis (CL) caused by *Leishmania donovani*, a pathogen more usually associated with visceral leishmaniasis, is now endemic in Sri Lanka. This report details the characteristics of 200 patients with locally acquired CL, who were recruited prospectively for an ongoing study into the genetic susceptibility to CL in Sri Lanka. In each case, the CL was confirmed by the demonstration of amastigotes in a direct smear and/or promastigotes in a culture. Although only 82% of the Sri Lankan population is Sinhalese, all 200 patients belonged to this ethnic group. The patients had a median age of 32 years (range=4-80 years). Most of them each had a single, non-tender, non-itching and dry lesion which had started as a papule and then gradually enlarged and ulcerated, with changes in the surrounding skin. None of the patients had any signs of systemic disease. Eleven (5.5%) each had at least one other affected family member. Patients with multiple lesions were most likely to be found in families with more than one affected member ( $P = 0.002$ ) but multiple lesions were not associated with diabetes mellitus ( $P > 0.05$ ). Although the results of passive detection under-estimate the true occurrence of a disease, the present data point towards enhanced susceptibility to CL among the Sinhalese and/or certain individuals, possibly determined by genetic factors.

## **Identification of geographically distributed sub-populations of *Leishmania* (*Leishmania*) major by microsatellite analysis.**



Al-Jawabreh A, Diezmann S, Mueller M, Wirth T, Schnur LF, Strelkova MV, Kovalenko DA, Razakov SA, Schwenkenbecher J, Kuhls K, Schoenian G.

BMC Evol Biol. 2008 Jun 24;8(1):183.

**ABSTRACT:** **BACKGROUND:** *Leishmania (Leishmania) major*, one of the agents causing cutaneous leishmaniasis (CL) in humans, is widely distributed in the Old World where different species of wild rodent and phlebotomine sand fly serve as animal reservoir hosts and vectors, respectively. Despite this, strains of *L. (L.) major* isolated from many different sources over many years have proved to be relatively uniform. To investigate the population structure of the species highly polymorphic microsatellite markers were employed for greater discrimination among its otherwise closely related strains, an approach applied successfully to other species of *Leishmania*. **RESULTS:** Multilocus Microsatellite Typing (MLMT) based on 10 different microsatellite markers was applied to 106 strains of *L. (L.) major* from different regions where it is endemic. On applying a Bayesian model-based approach, three main populations were identified, corresponding to three separate geographical regions: Central Asia (CA); the Middle East (ME); and Africa (AF). This was congruent with phylogenetic reconstructions based on genetic distances. Re-analysis separated each of the populations into two sub-populations. The two African sub-populations did not correlate well with strains' geographical origin. Strains falling into the sub-populations CA and ME did mostly group according to their place of isolation although some anomalies were seen, probably, owing to human migration. **CONCLUSION:** The model- and distance-based analyses of the microsatellite data exposed three main populations of *L. (L.) major*, Central Asia, the Middle East and Africa, each of which separated into two sub-populations. This probably correlates with the different species of rodent host.

## **INHIBITION BY DICATIONS OF IN VITRO GROWTH OF LEISHMANIA MAJOR AND LEISHMANIA TROPICA: CAUSATIVE AGENTS OF OLD WORLD CUTANEOUS LEISHMANIASIS.**

Rosypal A, Werbovets K, Salem M, Stephens C, Kumar A, Boykin D, Hall J, Tidwell R.

J Parasitol. 2008 Mar 11:1. [Epub ahead of print]

Old World cutaneous leishmaniasis is caused by infection with *Leishmania major* and *L. tropica*. Pentamidine and related dications exhibit broad spectrum antiprotozoal activity. Based on the previously reported efficacy of these compounds against related organisms, eighteen structural analogs of pentamidine were evaluated for in vitro antileishmanial activity, using pentamidine as the standard reference drug for comparison. Furan analogs and reversed amidine compounds were examined for activity against *L. major* and *L. tropica* promastigotes. The most active compounds against both *Leishmania* spp. were in the reversed amidine series. DB 745 and DB 746 exhibited the highest activity against *L. major* and DB 745 was the most active compound against *L. tropica*. Both of these compounds exhibited 50% inhibitory concentrations (IC<sub>50</sub>) below one nanomolar for *L. major*. Ten reversed amidines were also tested for their ability to inhibit growth in an axenic amastigote model. Nine of ten reversed amidine analogs were active at concentrations below one micromolar. These results justify further study of dicationic compounds as potential new agents for treating cutaneous leishmaniasis.

## **Neglected infections of poverty in the United States of america.**



Hotez PJ.

*Department of Microbiology, Immunology, and Tropical Medicine, The George Washington University and Sabin Vaccine Institute, Washington, D.C., United States of America.*

PLoS Negl Trop Dis. 2008 Jun 25;2(6):e256.

In the United States, there is a largely hidden burden of diseases caused by a group of chronic and debilitating parasitic, bacterial, and congenital infections known as the neglected infections of poverty. Like their neglected tropical disease counterparts in developing countries, the neglected infections of poverty in the US disproportionately affect impoverished and under-represented minority populations. The major neglected infections include the helminth infections, toxocariasis, strongyloidiasis, ascariasis, and cysticercosis; the intestinal protozoan infection trichomoniasis; some zoonotic bacterial infections, including leptospirosis; the vector-borne infections Chagas disease, leishmaniasis, trench fever, and dengue fever; and the congenital infections cytomegalovirus (CMV), toxoplasmosis, and syphilis. These diseases occur predominantly in people of color living in the Mississippi Delta and elsewhere in the American South, in disadvantaged urban areas, and in the US-Mexico borderlands, as well as in certain immigrant populations and disadvantaged white populations living in Appalachia. Preliminary disease burden estimates of the neglected infections of poverty indicate that tens of thousands, or in some cases, hundreds of thousands of poor Americans harbor these chronic infections, which represent some of the greatest health disparities in the United States. Specific policy recommendations include active surveillance (including newborn screening) to ascertain accurate population-based estimates of disease burden; epidemiological studies to determine the extent of autochthonous transmission of Chagas disease and other infections; mass or targeted treatments; vector control; and research and development for new control tools including improved diagnostics and accelerated development of a vaccine to prevent congenital CMV infection and congenital toxoplasmosis.

## **Leishmania infantum: tuning digitonin fractionation for comparative proteomic of the mitochondrial protein content.**

Hide M, Ritleng AS, Brizard JP, Monte-Allegre A, Sereno D.

*IRD de Montpellier, Génétique et Evolution des Maladies Infectieuses, IRD/CNRS (UMR 2724), 34394, Montpellier, France, mallorie.hide@ird.fr.*

Parasitol Res. 2008 Jun 25.

*Leishmania infantum* belongs to the Kinetoplastidae that is characterized by a specific mitochondrial DNA, the kinetoplast. This parasite is responsible for both benign cutaneous leishmaniasis and severe visceral leishmaniasis in humans. Molecular determinants of such differences in pathogenesis are not well understood, and the parasites as well as their hosts may contribute to the disease phenotype. Factors that help parasite to adapt its metabolism to nutritional conditions encountered in different location might play pivotal roles in controlling parasite development in these various host environments. Thus, we have decided to initiate studies aimed to compare the mitochondrial protein content of *L. infantum*. To avoid the drawback caused by the most abundant proteins such as tubulin and proteins of the cytoskeleton present in whole cell extract, we have decided to fractionate the subcellular components of the cells. Using both cytosolic and mitochondrial markers, we have improved a protein pre-fractionation protocol using digitonin that allowed us to generate an enriched mitochondrial fraction.



---

## Macrophage and T-Cell Gene Expression in a Model of Early Infection with the Protozoan *Leishmania chagasi*.

Ettinger NA, Wilson ME.

*Interdisciplinary Graduate Program in Molecular and Cellular Biology, University of Iowa, Iowa City, Iowa, United States of America.*

PLoS Negl Trop Dis. 2008 Jun 25;2(6):e252.

Visceral leishmaniasis is a potentially fatal infectious disease caused by the protozoan parasite *Leishmania infantum/chagasi* in the New World, or by *L. donovani* or *L. infantum/chagasi* in the Old World. Infection leads to a variety of outcomes ranging from asymptomatic infection to active disease, characterized by fevers, cachexia, hepatosplenomegaly and suppressed immune responses. We reasoned that events occurring during the initial few hours when the parasite encounters cells of the innate and adaptive immune systems are likely to influence the eventual immune response that develops. Therefore, we performed gene expression analysis using Affymetrix U133Plus2 microarray chips to investigate a model of early infection with human monocyte-derived macrophages (MDMs) challenged with wild-type *L. chagasi* parasites, with or without subsequent co-culture with *Leishmania*-naïve, autologous T-cells. Microarray data generated from total RNA were analyzed with software from the Bioconductor Project and functional clustering and pathway analysis were performed with DAVID and Gene Set Enrichment Analysis (GSEA), respectively. Many transcripts were down-regulated by infection in cultures containing macrophages alone, and the pattern indicated a lack of a classically activated phenotype. By contrast, the addition of autologous *Leishmania*-naïve T cells to infected macrophages resulted in a pattern of gene expression including many markers of type 1 immune cytokine activation (IFN- $\gamma$ , IL-6, IL-1 $\alpha$ , IL-1 $\beta$ ). There was simultaneous up-regulation of a few markers of immune modulation (IL-10 cytokine accumulation; TGF- $\beta$  Signaling Pathway). We suggest that the initial encounter between *L. chagasi* and cells of the innate and adaptive immune system stimulates primarily type 1 immune cytokine responses, despite a lack of classical macrophage activation. This local microenvironment at the site of parasite inoculation may determine the initial course of immune T-cell development.

## Accelerated Control of Visceral *Leishmania donovani* Infection in IL-6-Deficient Mice.

Murray HW.

*Department of Medicine, Weill Cornell Medical College, New York, NY 10065.*

Infect Immun. 2008 Jun 23.

In patients with visceral leishmaniasis, increased circulating levels of IL-6 regularly accompany fully-expressed, progressive infection (kala-azar). To experimentally test the role of IL-6, responses to intracellular *Leishmania donovani* infection were compared in livers of IL-6(-/-) vs. wild-type mice. IL-6(-/-) mice showed enhanced control of infection and earlier, rapid parasite killing along with additional evidence of a stimulated antileishmanial Th1 cell-type response: increased circulating IFN- $\gamma$ , accelerated granuloma assembly and heightened responsiveness to chemotherapy. In this model of visceral leishmaniasis, IL-6 appears to act in a suppressive, macrophage-deactivating fashion, identifying it as a potential target for therapeutic blockade.



---

## Leishmaniasis of the tongue due to leishmania infantum.

Laguna EV, Martinez AA, Cogolludo EF, Martín L, Merano F, Valdés MA.

*Hospital Severo Ochoa Leganés, Avd/Orellana s/n, 28911 Leganés, Madrid, Spain.*

Eur J Dermatol. 2008 Jun 23;18(4):472-473. [Epub ahead of print]

## Imported tropical infectious ulcers in travelers.

Zeegelaar JE, Faber WR.

*Department of Dermatology, Academic Medical Centre, Amsterdam, the Netherlands*  
*Department of Dermatology, Flevoziekenhuis, Almere, the Netherlands.*

Am J Clin Dermatol. 2008;9(4):219-32.

Skin ulcers are a commonly encountered problem at departments of tropical dermatology in the Western world. Furthermore, the general dermatologist is likely to be consulted more often for imported chronic skin ulcers because of the ever-increasing travel to and from tropical countries. The most common cause of chronic ulceration throughout the world is probably pyoderma. However, in some parts of the world, cutaneous leishmaniasis is one of the most prevalent causes. Mycobacterium ulcerans is an important cause of chronic ulcers in West Africa. Bacterial infections include pyoderma, mycobacterial infections, diphtheria, and anthrax. Pyoderma is caused by Staphylococcus aureus and/or beta-hemolytic streptococci group A. This condition is a common cause of ulcerative skin lesions in tropical countries and is often encountered as a secondary infection in travelers. The diagnosis is often made on clinical grounds. Antibacterial treatment for pyoderma should preferably be based on culture outcome. Floxacillin is generally active against S. aureus and beta-hemolytic streptococci. Infection with Mycobacterium ulcerans, M. marinum, and M. tuberculosis may cause ulcers. Buruli ulcers, which are caused by M. ulcerans, are endemic in foci in West Africa and have been reported as an imported disease in the Western world. Treatment is generally surgical, although a combination of rifampin (rifampicin) and streptomycin may be effective in the early stage. M. marinum causes occasional ulcerating lesions in humans. Treatment regimens consist of combinations containing clarithromycin, rifampin, or ethambutol. Cutaneous tuberculosis is rare in travelers but may be encountered in immigrants from developing countries. Treatment is with multiple drug regimens consisting of isoniazid, ethambutol, pyrazinamide, and rifampin. Cutaneous diphtheria is still endemic in many tropical countries. Cutaneous diphtheria ulcers are nonspecific and erythromycin and penicillin are both effective antibacterials. Antitoxin should be administered intramuscularly in suspected cases. Anthrax is caused by spore-forming Bacillus anthracis. This infection is still endemic in many tropical countries. Eschar formation, which sloughs and leaves behind a shallow ulcer at the site of inoculation, characterizes cutaneous anthrax. Penicillin and doxycycline are effective antibacterials. Cutaneous leishmaniasis is caused by different species belonging to the genus Leishmania. The disorder is one of the ten most frequent causes of skin diseases in travelers returning from (sub)tropical countries. The clinical picture is diverse, ranging from a painless papule or nodule to an ulcer with or without a scab. Treatment depends on the clinical manifestations and the species involved. Sporotrichosis, chromo(blasto)mycosis, and mycetoma are the most common mycoses that may be accompanied by ulceration. Infections are restricted to certain regions and often result from direct penetration of the fungus into the skin. Anti-mycotic treatment depends on the microorganism involved. The most common causes of infectious skin ulceration



encountered in patients from tropical countries who present at a department of tropical dermatology are reviewed in this article.

## **Cutaneous leishmaniasis of the lower lip in a United States soldier.**

Roundy S, Almony J, Zislis T.

*Womack Army Medical Center Oral & Maxillofacial Surgery Service, Ft Bragg, NC 28310, USA.  
shad.roundy@us.army.mil*

J Oral Maxillofac Surg. 2008 Jul;66(7):1513-5.

## **Validation of a recombinant based antibody ELISA for diagnosis of human and canine leishmaniasis.**

Daprà F, Scalone A, Mignone W, Ferroglio E, Mannelli A, Biglino A, Zanatta R, Gradoni L, Rosati S.

*Department of Animal Production, Epidemiology and Ecology, Faculty of Veterinary Medicine, University of Torino, Via Leonardo da Vinci 44, 10095 Grugliasco (TO), Italy.*

J Immunoassay Immunochem. 2008;29(3):244-56.

In this study, a recombinant chimeric antigen (CA) ELISA was validated as a single test for both human and dog leishmaniasis. Serum panels included 327 human and 339 canine IFAT-positive and 1113 human and 1078 canine IFAT-negative samples. CA-ELISA was carried out using the same serum dilution, and labelled protein A as secondary reagent. Test performances were calculated using ROC analysis. For the human panel, the test showed diagnostic accuracy (DA) 0.974, specificity (Sp) 97.12%, sensitivity (Se) 91.44%, and concordance (K) 0.88. The dog panel showed DA 0.998, Sp 99.54%, Se 98.54%, and K 0.98. The proposed method is the best recombinant antigen-based ELISA, and can be used as IFAT substitute for mass screening.



## Political and regulatory

### The Right to Health

*The Right to Health* is published by Office of the United Nations High Commissioner for Human Rights (OHCHR), in collaboration with the World Health Organization, in an effort to explore the links between health care and human rights. The document begins by explaining what the right to health is. It illustrates its implications for specific individuals and groups, and then elaborates upon the obligations of States to ensure that the right to health is met. Specific issues highlighted include a consideration of how the right to health applies to women, children, migrants and people living with HIV/AIDS. *The Right to Health* ends with an overview of national, regional and international accountability and monitoring mechanisms.

Fact sheet available at: <http://www.ohchr.org/Documents/Publications/Factsheet31.pdf>

### **FIND celebrates five years of its successful program to combat neglected diseases with more efficient diagnostic tests**

Launched at a meeting of the World Health Assembly in 2003 with a grant from the Bill and Melinda Gates Foundation, FIND (Foundation for Innovative New Diagnostics) has become an influential leader in the development of new diagnostic tests to identify tuberculosis (TB), especially in patients with the multidrug-resistant (MDR) and extensively drug-resistant (XDR) forms of the disease. Drug resistant cases are currently rising in various parts of the world and XDR TB is generally incurable. The exact spread of drug-resistant tuberculosis remains largely unknown due to the fact that only a few countries are equipped to properly diagnose it.

Read more at: [http://www.finddiagnostics.org/news/press/five\\_years\\_jun08.shtml](http://www.finddiagnostics.org/news/press/five_years_jun08.shtml)