**Title:** New tools for monitoring drug resistance and treatment response in Visceral Leishmaniasis in the Indian subcontinent
**Acronym:** Kaladrug-R
**Contract/Grant agreement number:** 222895
**EC contribution:** 3.000.000 €
**Duration:** 48 months
**Starting date:** targeted, second semester 2008
**Instrument:** Medium-scale focused research project

**Summary:** This project concerns the effective control of a most neglected parasitic disease, Kala-Azar. By providing knowledge and tools relevant for monitoring the effectiveness of the existing few drugs, we will contribute to safeguard them and establish the bases for their longer-term and more rational use. We will use an integrated and multidisciplinary approach to develop new tools for monitoring response to treatment and drug resistance. Our clinical and epidemiological work should contribute to an early identification of the persons at higher risk of treatment failure and a more rational therapeutic attitude. Molecular mechanisms leading to drug resistance will be deciphered by an approach gathering cell and molecular biology (including genomics) together with biochemistry. Tools for the detection of drug resistance will be developed and they will be simplified as much as possible to be applicable and affordable in the endemic countries. The epidemiological dynamics of drug resistance will be modelled and strategies for maximizing intervention success will be recommended. Last but not least, a particular attention will be paid to technology transfer and training activities.

**Problem:** Visceral leishmaniasis (VL) or Kala-Azar (KA) has been reported from 51 countries around the world with an annual incidence of 500,000 cases, about 90% of which occur in India, Nepal, Bangladesh, Sudan and Brazil. Early treatment is a major pillar of the current KA elimination programme, which was launched by the governments of India, Nepal and Bangladesh. The arsenal of available drugs is limited. Because of its toxicity and because of emerging drug resistance, the former first-line drug sodium stibogluconate (SSG) was recently replaced by miltefosine (MIL) in the Indian subcontinent, but MIL is an oral drug with a long half-life, and it is feared that resistance will rapidly emerge. Wide-spread over-the-counter drug sales and irrational drug prescribing further increase this risk. Moreover, the region is confronted with an expanding HIV-epidemic, and we expect to see more HIV-VL co-infections which will generate major therapeutic challenges. Combination regimens for VL are under clinical development, but the drug policy will take several more years to change. Meanwhile, the effectiveness of current drugs needs to be safeguarded in order to cure patients and to sustain the control of VL. For this, the uninterrupted supply of quality drugs, the promotion of treatment adherence and the monitoring of treatment effectiveness and of drug resistance will be pivotal. There is a direct need for new tools to allow monitoring treatment effectiveness and drug resistance because (i) validated methods to monitor treatment effectiveness under routine conditions do not exist, (ii) there are discrepancies in assays for the assessment of drug resistance in *Leishmania* parasites, (iii) the knowledge on mechanisms of emergence of drug resistance, its dynamics and the impact of the introduction of new drugs is poor, and (iv) molecular tools for high throughput monitoring of drug resistance do not exist. Clinical and laboratory research is urgently needed to support the drug policy of the VL elimination programme.

**Aim:** develop, evaluate and disseminate new tools for evaluation of drug resistance in *L. donovani* as well as innovative methodologies for monitoring Kala-Azar treatment effectiveness in routine conditions.

**(Expected) results:** (1) An innovative approach for monitoring effectiveness of VL treatment in the health services of *L. donovani* endemic areas and in routine conditions. (2) Comprehensive collections of well-documented clinical samples and parasites including all available epidemiological information. (3) A reference genome sequence of *L. donovani* from the region that will serve as reference for identifying markers of drug resistance and SNPs for population genetic studies. (4) Upgraded and standardized assays for testing *in vitro* susceptibility against antileishmanial drugs. (5) Deciphering molecular mechanisms underlying natural resistance against SSG and MIL and clarify the relation between natural resistance and clinical treatment failure. (6) Validated simple molecular techniques for monitoring natural resistance to SSG and MIL. (7) Molecular mechanisms of experimental resistance against PMM and markers. (8) Reference centers for monitoring the effectiveness of VL treatment and drug resistance and platforms for translating knowledge into policy. (9) Mathematical model for the emergence and spread of drug resistance, including the role of asymptomatic infections and allowing for combined intervention strategies. (10) Highly variable DNA markers for epidemiological and population studies. (11) Real-time evidence to adapt VL drug policy in a flexible way to evolving drug resistance patterns. (12) Expertise and capacity for *Leishmania* genome variation analysis for clinical/epidemiological trait association studies

**Potential applications:** Our clinical and epidemiological work should contribute to (i) an early identification (in clinical settings as well as in peripheral health centres) of the persons at higher risk of treatment failure and (ii) a more rational therapeutic attitude. This would have a direct impact on treatment costs. By deciphering the molecular mechanisms leading to drug resistance and providing the respective detection tools and by understanding the epidemiological dynamics of its emergence and spreading, our consortium will support control programmes relying on chemotherapy (among other methods), like the Kala-azar elimination programme.

**Project web-site:** a special Kaladrug-R page will be created in our networking platform www.leishrisk.net

**Key words:** visceral leishmaniasis, Indian sub-continent, treatment effectiveness, drug resistance, miltefosine, antimonials, paromomycine, comparative genomics

**Coordinator:** Jean-Claude Dujardin, Institute of Tropical Medicine, Nationalestraat 155, B-2000 Antwerpen, Belgium; email: jcdujardin@itg.be

**Partners:** Graham Coombs, Strathclyde University, Glasgow, UK, graham.coombs@strath.ac.uk; Louis Maes, University of Antwerp, Belgium, louis.maes@ua.ac.be; Gabrielle Schönian, Universitaetsmedizin Charité Berlin, Germany, gabriele.schoenian@charite.de; Martin Eichner, University of Tuebingen, Germany, martin.eichner@uni-tuebingen.de; Matt Berriman, Wellcome Trust Sanger Institute, UK, mb4@sanger.ac.uk; Shyam Sundar, Banaras Hindu University, Varanasi, India, doctorshyamsundar@gmail.com; Suman Rijal, B.P. Koirala Institute of Health Sciences, Dharan, Nepal, sumanrijal2@yahoo.com; Poonam Salotra, Institute of Pathology (ICMR), Safdarjung Hospital Campus, New Delhi, India, salotra@del1.vsnl.net.in; Syamal Roy, Indian Institute Chemical Biology (CSIR), Calcutta, India, sroy@iicb.res.in