YALE JOURNAL OF BIOLOGY AND MEDICINE 94 (2021), pp.351-360.

PERSPECTIVES

YALE JOURNAL OF BIOLOGY AND MEDICINE
YJBM
CELEBRATING 90 YEARS 1928–2018

Ending the Neglect of Treatable Bacterial Zoonoses Responsible for Non-Malaria Fevers

Cyrille Goarant[a], Koussay Dellagi[b], and Mathieu Picardeau[c,*]

[a]*Institut Pasteur de Nouvelle-Calédonie, Unité de Recherche et d'Expertise sur la Leptospirose, Nouméa, New Caledonia;* [b]*Institut Pasteur, Direction Internationale, Paris, France;* [c]*Institut Pasteur, Unité Biologie des Spirochètes, Paris, France*

Bacterial zoonotic diseases such as leptospirosis, Q fever, melioidosis, spotted fever group rickettsioses, and brucellosis are increasingly recognized causes of non-malaria acute fevers. However, though readily treatable with antibiotics, these diseases are commonly misdiagnosed resulting in poor outcomes in patients. There is a considerable deficit in the understanding of basic aspects of the epidemiology of these neglected diseases and diagnostic tests for these zoonotic bacterial pathogens are not always available in resource-poor settings. Raising awareness about these emerging bacterial zoonoses is directly beneficial to the patients by allowing a test-and-treat approach and is essential to control these life-threatening diseases.

INTRODUCTION

Historically, malaria was considered the most common infectious cause of fever in tropical countries. In 2018, an estimated 228 million cases of malaria occurred worldwide, mostly in Africa (93% of cases), and there were an estimated 405,000 deaths from malaria globally [1]. Arboviral infections such as dengue are also common causes of acute febrile illness in tropical countries with more than one third of the world's population living in areas at risk for infection [2]. However, significant over-diagnosis of malaria in different parts of Africa and Asia has been recently reported. Similarly, clinical suspicion overestimates the true number of dengue patients [3,4]. Studies have shown that bacterial zoonoses such as leptospirosis, Q fever, melioidosis, spotted fever group rickettsioses, and brucellosis are major causes of non-malarial febrile illness [5-14]. Most of these bacterial infections are considered as emerging and neglected diseases and have been much less investigated than malaria or viral illnesses.

As identification of a bacterial etiology opens avenues to the administration of lifesaving antibiotics, a "test-and-treat" approach targeting bacterial infections causing endemic and/or epidemic febrile illness is directly beneficial to patients. On the contrary, if untreated, acute fever of bacterial origin can progress to cause multiple organ failure and death. However, these bacterial infections are often undiagnosed or misdiagnosed as a result of non-specific clinical manifestations, lack of specific and sensitive diagnostic tests, and low awareness amongst clinicians.

Bacterial zoonoses not only affect human health, but also livestock farming and agricultural development. Zoonotic diseases, such as brucellosis and leptospirosis can lead to infertility, loss of milk, and abortion in live-

*To whom all correspondence should be addressed: Mathieu Picardeau, Institut Pasteur, Paris, France; Email: mpicard@pasteur.fr; ORCID iD: 0000-0002-5338-5579.

Abbreviations: DALYs, disability-adjusted life years.

Keywords: neglected tropical diseases, leptospirosis, tropical countries, antibiotics

Table 1. Priority neglected zoonotic diseases according to WHO.

Disease	Pathogen
Buruli ulcer	Bacteria
Chagas disease	Parasite
Dengue and Chikungunya	Virus
Dracunculiasis (guinea-worm disease)	Parasite
Echinococcosis	Parasite
Foodborne trematodiases	Parasite
Human African trypanosomiasis (sleeping sickness)	Parasite
Leishmaniasis	Parasite
Leprosy (Hansen's disease)	Bacteria
Lymphatic filariasis	Parasite
Mycetoma, chromoblastomycosis and other deep mycoses	Fungi
Onchocerciasis (river blindness)	Parasite
Rabies	Virus
Scabies and other ectoparasites	Parasite
Schistosomiasis	Parasite
Soil-transmitted helminthiases	Parasite
Snakebite envenoming	other
Taeniasis/Cysticercosis	Parasite
Trachoma	Bacteria
Yaws (Endemic treponematoses)	Bacteria

In 2013, the 66th World Health Assembly established a list of selected neglected tropical diseases that could be targeted to improve the health and social well-being of affected populations [68]. The list was updated in 2017 with the addition of chromoblastomycosis and other deep mycoses, scabies and other ectoparasites and snakebite envenoming [69].

stock, causing great economic or subsistence-resources losses. For example, economic burden of brucellosis, for which the main symptom in cattle is repetitive abortions, was estimated at US$60 million per year when the prevalence was around 5% in Argentina, and in Nigeria losses were estimated at US$575,605 (prevalence 7% to 12%) [15]. In New Zealand, the cost of human and animal leptospirosis was estimated at US$18.80 million a year [16].

No bacterial zoonotic disease is included in the list of 20 neglected tropical diseases identified by the World Health Organization (WHO) (Table 1). Leptospirosis, rickettsioses, and other bacterial zoonotic diseases are lacking the advocacy required to mobilize political support and funding from non-governmental agencies (NGOs) for their control in endemic countries [17]. When comparing global investment levels on the basis of burden of disease by disability-adjusted life years (DALYs) across 18 infectious diseases, including neglected diseases such as dengue, leptospirosis was ranked last [18]. Funding for malaria research from 2000 to 2017 was US$5.6 billion, 80 times that for leptospirosis (US$0.07 billion) [19,20] (Table 2).

A better understanding of the local epidemiology of zoonotic pathogens contributing to the burden of febrile diseases is essential for clinicians and diagnostic laboratories. More importantly discriminating between bacterial infections and other causes of fever is of great importance to triage patients in need of antibiotics. One Health approaches are also particularly relevant for the management, prevention, and control of bacterial zoonoses.

BACTERIA CAUSING NON-MALARIAL FEBRILE ILLNESSES

Many studies have shown the importance of conducting surveys on the etiologies of acute febrile illnesses to identify regional and seasonal specificities and the changing patterns of the different etiologies, especially in the context of a decreasing incidence of malaria. For example, the diagnosis of inpatients admitted with fever in Tanzania between 2007 and 2008 was only of 1.6% for malaria, while bacterial zoonoses were identified among 26.2% patients; 13.6% had brucellosis, 33.9%

Table 2. The burden of malaria and neglected tropical diseases expressed in disability-adjusted life years (DALYs).

Disease	DALY per 100,000	Reference
Malaria	897.6 (728.1-1094.8)	[70]
Melioidosis	84 (57-120)	[35]
Cholera	65 (49–84)	[71]
Leishmaniasis	58.6 (48.2-69.7)	[70]
Schistosomiasis	42.1 (23.3-77.8)	[70]
Leptospirosis	42 (18.1-66)	[72]
Lymphatic filariasis	28.9 (15.7-47.1)	[70]
Rabies	17.3 (12.7-21.2)	[70]
Dengue	15.8 (10.1-27.4)	[70]
Scrub typhus	13*	[45]

*2012, Laiwu, China

had leptospirosis, 20.3% had Q fever, 30.5% had spotted fever group rickettsioses and 1.8% had typhus group rickettsioses [7]. A similar study in the same location has shown that the leptospirosis incidence has dropped from 75-102/100,000 people during 2007-2008 to 8-11 cases/100,000 people during 2012-2014 [21], while the incidence of brucellosis remains stable to 33-35/100,000 people [22]. In a recent review on zoonotic causes of fever in malaria-endemic countries, the majority of zoonoses (17 out of 30) were identified as bacteria and *Leptospira* and non-typhoidal *Salmonella* were the most frequently reported pathogens [13].

EXAMPLES OF NEGLECTED BACTERIAL PATHOGENS

Febrile illness can be attributable to many bacterial zoonotic pathogens (Table 3). Zoonotic pathogens such as *Coxiella burnetii* (Q fever) and *Bartonella henselae* (cat scratch fever) [13] will not be considered here because only limited evidence is available for their contribution to the burden of febrile illnesses in communities. The agent of salmonellosis and other foodborne zoonoses, common in both developing and industrialized countries and bacterial pathogens occurring in only few locations (ie, plague) will also not be considered here.

Leptospira (Leptospirosis)

Leptospirosis is a zoonosis of global distribution frequently considered to emerge or re-emerge [23,24]. Annually, an estimated 1.03 million cases of leptospirosis lead to almost 60,000 deaths worldwide, a global burden in the same range as schistosomiasis, leishmaniasis, or lymphatic filariasis [25] (Table 2).

Pathogenic *Leptospira* can virtually infect any mammal. The natural or maintenance hosts usually have no or mild symptoms after infection. In contrast, accidental hosts such as humans may develop severe, sometimes even lethal, disease. In many mammals, leptospirosis is a reproductive disease associated with fetal deaths and abortions [26]. Virulent leptospires reside in the kidney tubules of reservoir animals and are excreted within the urines in the environment where they are assumed to only survive, but not multiply [27]. In humans, infections mostly occur through exposure to contaminated water and soil environments, during both occupational and recreational activities. Due to the non-specific clinical presentation, biological confirmation is required, but tests are not always available, and they have been reported with varying diagnostic performances in distinct countries [28,29]. Medical treatment relies on first-line antibiotics like amoxicillin, erythromycin, or 3rd generation cephalosporins that should be given in the first days of symptoms upon suspicion and without awaiting the results of laboratories [30]. Vaccination is available for animals and humans but the inactivated whole-cell vaccines confer short term protection and do not cross-protect against the large number of pathogenic serovars [31]. There is reasonable evidence that animal vaccination has successfully decreased the burden of human leptospirosis in New Zealand [32]. Prevention therefore mostly relies on personal protective equipment and general hygiene together with increased awareness in at-risk populations.

Burkholderia (Melioidosis)

Melioidosis is an environment-borne bacterial disease caused by *Burkholderia pseudomallei* with the highest burden in Asia (Thailand) and the Pacific (Australia) [33], but the disease is also increasingly recognized throughout the tropics in both the African and American

Table 3. Selected bacterial zoonoses.

Zoonotic disease	Causative pathogen	Region	Main reservoirs	Mode of transmission to humans	Treatment (ATB)
Brucellosis	*Brucella* spp.	Worldwide	Cattle, goats, sheep, pigs, and dogs; marine mammals (*B. pinnipedialis* and *B. ceti*)	Ingestion of unpasteurized dairy products or undercooked meat, contact with mucous membranes and broken skin	Doxycycline + rifampicin
Ehrlichiosis	*Ehrlichia* spp.	Southeastern and south-central United States	Sheep, cattle, deer, dogs, and cats	Tick bite	Doxycycline
Leptospirosis	*Leptospira* spp.	Worldwide	Rodents, cattle, dogs, pigs, water, and soil	Contact of mucous membranes, and broken skin with surface water or soil contaminated with urine of infected animals	Doxycycline, penicillin or cephalosporins
Murine typhus	*Rickettsia prowazekii* and *Rickettsia typhi*	Temperate countries	Rats	Contact with flea infected feces or flea bite	Doxycycline
Melioidosis	*Burkholderia* spp.	Southeast Asia and northern Australia	Sheep, goats, and pigs, soil and water	Percutaneous inoculation, contamination of wounds, ingestion of soil or contaminated carcasses, or inhalation	Trimethoprim-sulfamethoxazole or Amoxicillin/ clavulanic acid
Spotted Fever	*Rickettsia rickettsii, Rickettsia conorii, Rickettsia africae*, etc	United States/ Europe/ Africa	Dogs	Tick bite	Doxycycline
Anaplasmosis	*Anaplasma phagocytophilum*	Upper midwestern and northeastern United States/ Northern and central Europe	Cattle, sheep, goats	Tick bite	Doxycycline
Q fever	*Coxiella burnetii*	Worldwide	Sheep, goats, and cattle	Inhalation of aerosols	Doxycycline
Cat scratch fever	*Bartonella henselae*	Worldwide	Cats	Scratch, bite or by infected saliva through broken skin	Azithromycin
Scrub typhus	*Orientia tsutsugamushi*	Asia and Australasia	Rats	Mite (“chigger”) bite	Doxycycline

continents [33]. Most cases are thought to occur in rural locations with limited access to diagnosis [34]. Melioidosis is considered to cause 165,000 cases annually, with as many as 89,000 deaths, illustrating its high fatality rate [33]. Its global burden in terms of DALYs is relatively high [35] (Table 2). Although zoonotic, melioidosis in humans mostly originates in water and soil; direct infection from an animal source is likely rare [34]. Similarly

to leptospirosis [27], the incidence of melioidosis is under strong influence of the weather and climate, with heavy rain caused by the monsoon or cyclones triggering exposure to the pathogen [36,37]. Humans and animals are infected through skin or respiratory contact with water, water droplets, or soils contaminated with *B. pseudomallei*. Most infections are spontaneously cleared by the immune response and only evidenced by serology. People with diabetes are more severely affected by infections [34]. Another subset of infected people (ca. 4%) get into a latent asymptomatic infection, with possible clinical manifestations months to years after infection [38]. Patients frequently present with pneumonia, with or without bacteremia, sometimes evolving towards septic shock. Clearing the pathogen from an infected body requires specific and prolonged antimicrobial therapies, notably because of the ability of the bacteria to survive inside macrophages [34]. Due to high level of exposure to *B. pseudomallei* and other closely related species, the serological diagnosis is of very small utility. Therefore, the preferred biological confirmation relies on culture isolation, whereas molecular detection is rarely used [34]. Although *B. pseudomallei* is classified as a potential bioterrorism agent and imposes a high burden in endemic areas, there has been no vaccine candidate registered or even reaching clinical trials in humans [34]. Besides personal protective equipment and behavior, especially for people with diabetes, there has not been strong progress made in the prevention and control of melioidosis [39,40].

Scrub Typhus and Other Rickettsiales Infections

Pathogens in the genera *Orientia*, *Rickettsia*, *Ehrlichia*, and *Anaplasma* are obligate intracellular bacteria in the Order Rickettsiales and Families Rickettsiaceae and Anaplasmataceae. All of these Rickettsiales have at least part of their lifecycle in arthropod vectors such as ticks (spotted fever, anaplasmosis, and ehrlichiosis), fleas (murine typhus), or mites (scrub typhus) (Table 3). Spotted fever group rickettsioses, typhus group rickettsioses, and human granulocytic anaplasmosis are causes of acute febrile illnesses worldwide. Human monocytic ehrlichiosis also results in acute febrile illness across the Americas, whereas scrub typhus occurs mostly in Asia and Australasia, even though cases have been recently identified in the Middle East, Africa, and South America suggesting that this disease may be more widespread than previously appreciated [41]. For most rickettsial infections, poor outcomes can occur without early identification and specific antibiotic treatment, usually with doxycycline. There is no vaccine available for any rickettsial infections including scrub typhus. Prevention mostly relies on avoiding contact with arthropod vectors by changing agricultural practices, for example.

Emerging infections with new *Ehrlichia* and *Anaplasma* species have become more frequently diagnosed as the cause of human infections, as animal reservoirs and tick vectors have increased in numbers and humans have inhabited areas where reservoir and tick populations are high [41]. Although ehrlichiosis, anaplasmosis, murine typhus, and spotted fever are globally emerging, a better burden estimate of these causes of acute fever is needed and only scrub typhus will be considered here. Scrub typhus (caused by *Orientia tsutsugamushi*) has been considered the most important rickettsiosis in densely populated countries in Asia [42,43]. It was estimated that there are one million cases each year [43] and mortality rates range from 1.5% if treated to 6% if untreated [44]. A single study from eastern China estimated the average DALYs at 9 between 2006 to 2012 with a constant increase over the years [45] (Table 2).

Brucella (Brucellosis)

Caused by various *Brucella* species (mostly *Brucella melitensis*), brucellosis is one of the most common bacterial zoonotic diseases affecting cattle, swine, goats, sheep, and causing over 500,000 human cases throughout the world every year but the burden is probably largely underestimated and far higher [5,46]. In the autonomous region of Inner Mongolia, China, the incidence of human brucellosis was estimated to be close to 300,000 new cases from 2010 to 2014 [46]. Exposure to a *Brucella*-contaminated environment, mainly through the ingestion of unpasteurized dairy products and direct contact with infected animals, results in non-distinct acute or chronic febrile illness in humans, usually not fatal. Although livestock vaccination against brucellosis has been effective in reducing the burden of disease in many parts of the world [5], the current vaccines cannot protect against all *Brucella* species. Doxycycline in combination with rifampicin remains the most common treatment of brucellosis in humans. Successful reduction of human brucellosis has been obtained by the systematic pasteurization of milk and milk products combined with animal vaccination programs [47].

FUTURE DIRECTIONS

Diagnostic Tools

The bacterial zoonotic pathogens described above are notoriously difficult to diagnose either because the early symptoms and signs are nonspecific, often mimicking malaria or viral illnesses, or rapid diagnostic tests are not available in resource-poor settings where the highest burden occurs. Delay in the diagnosis or misdiagnosis can lead to fatal outcomes stressing the need for developing rapid point of care diagnostic tests particularly for re-

source-poor countries. Distinguishing between infections of bacterial and viral etiology by pathogen detection (see below) or identification of specific biomarkers will be particularly important for antibiotic treatment decisions. Patients are rarely tested for the full range of pathogens responsible for acute fever. New diagnostic assays that can detect infections with the full range of febrile illness agents are urgently needed [48]. Multiplex PCR assays or multiplex immunoassays would be highly valuable to screen populations with acute febrile illnesses. Multiplex PCR assays targeting specific genes of *Orientia tsutsugamushi*, *Rickettsia typhi*, and *Leptospira interrogans* [49] or chikungunya virus, dengue virus, and pathogenic *Leptospira* [50] or malaria, dengue virus, and *Leptospira* [51] have thus been described. A new high-throughput virus-detection assay based on microfluidic PCRs being able to detect a wide range of viruses in thousands of collected mosquitoes or ticks was recently developed [42,43]. A similar high-throughput chip system capable of detecting bacterial zoonotic pathogens in one blood sample would be particularly useful. Primers and probes specifically targeting bacterial zoonotic pathogens and viruses could be used to perform large scale epidemiological survey screening of patients with acute febrile syndrome; considering that for each pathogen, the detectable load in blood may depend on the time of sampling (days after illness onset). A mobile suitcase laboratory containing all equipment and reagents for performing isothermal amplification assays of different pathogens in field studies can be used [52,53]. As antibodies can take more than 1 week to be reliably detected, serology may not be appropriate for early diagnosis and initiation of adequate treatment. However, antibody-based assays are informative in terms of understanding the exposure of populations to different pathogens. A multiplex immunoassay with key specific immunodominant epitopes was designed to discriminate antibody responses (both IgM and IgG) to eight tick-borne pathogens such as *Anaplasma phagocytophilum* (human granulocytic anaplasmosis) and *Rickettsia rickettsii* (Rocky Mountain spotted fever) [42].

For assays described above, the instrument used can be costly and not available at an affordable price to most of the world's lower income countries. It is recommended to identify reference laboratories where this technology could be developed but the major challenge remains the development of new low-cost diagnostic tests to support large epidemiological studies.

Clinical Surveys and Syndromic Approach

In contrast to infections that cause diarrhea and pneumonia [54,55], we lack true estimates of infections that cause unspecified febrile illnesses. The information gained from syndrome surveillance will be of importance for clinicians but also for prioritizing resource allocations and identifying potential preventive measures (see below). Clinical surveys targeting patients with acute febrile illness visiting in hospitals or admitted in intensive care units should be best conducted along a whole year to cover all seasons. Furthermore and in order to optimize the medical gain from the investigation and to cover wider diagnostic fields, the cohort of febrile patients should be investigated for broader range of possible etiologies including bacteria (in the frame of a composite diagnostic package targeting treatable life-threatening acute bacterial infections such as leptospirosis, brucellosis, melioidosis, ricketsiosis, borreliosis, gram negative sepsis, etc.). A recent meta-analysis showed that there is a lack of consensus on definitions of cases and study design, heterogeneity in the tested pathogens and adequate methods of data analysis [56]. There is a need for new studies with stringent epidemiological and diagnostic criteria.

One Health and Interventions

One health or "the collaborative efforts of multiple disciplines working locally, nationally, and globally, to attain optimal health for people, animals and our environment" has been defined almost two decades ago as "a new imperative" [57]. Bacterial zoonoses, reaching humans through arthropod vectors, food of animal origin, direct or indirect contact with infected animals or acquired by humans and animals in the same environment, should be considered into the One Health conceptual framework. There are striking examples illustrating the benefits of this concept for human health. Among the most evident is the use of animal vaccination to avoid pathogen spillover to humans that notably allowed the quasi-eradication of human rabies in Europe through the vaccination of domestic, but also wild carnivores [58]. There is also evidence that animal vaccination can decrease the burden of human leptospirosis as described above [32]. Other animal-based strategies are based on multiple interventions, including "test-and-treat," livestock culling, sometimes combined with vaccination, which is largely used in the control of brucellosis in ruminants [59]. Combined with improved sanitation of milk and milk products, these animal-oriented strategies have resulted in major improvements in human health [60]. In arthropod-borne zoonotic diseases, a better knowledge of the biology and ecology of both the host and the vector proves useful to help delineate the infection risk. This is especially important for pathogens like *Rickettsia* and *Orientia* for which no vaccine is available [43]. The ecological modeling can notably help target control and prevention strategies which include reservoir or vector control [61,62]. Lastly, in some zoonoses, soil and water act as the reservoir where both humans and animals get infected (eg, melioidosis) or as a "secondary passive" reservoir (eg, leptospirosis), a better knowledge of the ecological conditions of the pathogen

development or survival is needed to better comprehend the risk. Studies focusing on understanding the ecological niche [33] or the lifestyle and habitat of the pathogen [27,63] are of prime importance to inform public health prevention and interventions.

Lobbying and Policy Changes

All disease control measures require a substantial amount of financial support that is usually unavailable for the developing countries. Bacterial zoonoses are not yet recognized as neglected diseases by the WHO, despite data that illustrate their high incidence and prevalence and the potential morbidity and costs to human life and livestock farming. Of the 20 diseases on WHO's list of NTDs, only five are zoonoses: *Taenia solium,* cysticercosis, echinococcosis, leishmaniasis, rabies, and human African trypanosomiasis (HAT). None are bacterial zoonoses. Including these zoonotic pathogens in the WHO list of NTDs would facilitate funding to implement zoonoses control programs [64]. A better disease burden estimate of causes of fever in developing countries would be most useful for policy and decision makers. Future goals should therefore include the evaluation of the impact of zoonoses by assessing parameters such as disease prevalence, incidence, morbidity, mortality, and economic loss. This should allow to estimate the burden and compare with other infections and identify the priority animal diseases to be tackled and to conceive effective diagnostic and preventive measures against these bacterial zoonotic infections.

A critical aspect for disease surveillance and risk assessment will be to ensure results comparability across territories, contrasted ecosystems and populations to allow data integration that generates a meaningful interpretation of the infectious risk at the regional and global scales. To this end, shared multi-partner protocols are warranted using similar case definitions, diagnostic tests with recognized performance and well-identified target populations as recently typified by the project FIEBRE [65]. In this regard, international networks of research institutions that have developed a tradition of collaboration and sharing are in a privileged position to structure such systemic approaches. This is the case of the Institut Pasteur International Network (IPIN) which brings together 32 institutions conducting research and surveillance on infectious diseases in extremely diverse ecosystems distributed over the five continents and which has included in its strategic scientific plan this vision for multidisciplinary and multipartner coordinated actions [66].

CONCLUSION

Despite causing relatively high mortality and morbidity, bacterial zoonotic diseases causing human febrile illness are poorly recognized even among the so-called "neglected tropical diseases." A major problem in tackling these bacterial zoonoses is that they are often difficult to diagnose and are frequently misdiagnosed. However, these bacterial zoonoses are treatable with antibiotics and many of them are entirely or largely preventable through One Health strategies. Future work, for example, needs to better evaluate the human health benefits of vaccination of cattle against brucellosis or leptospirosis.

Lobbying to overcome the neglect imposed on these diseases is challenging and this is particularly true as it is still often done on an individual disease basis. To increase the attention of a wide range of stakeholders including the media, politicians, philanthropists, and the general public, collecting together a group of diseases, here the life-threatening treatable bacterial zoonotic diseases causing febrile illness, will certainly add political and economic weight. The implementation of a strengthening surveillance of a group of zoonotic pathogens at the national level as done in Ethiopia [67], instead of individual programs for each pathogen, would also have maximum benefit on the economy and health of people and animals by increasing both awareness of zoonotic diseases and interactions between veterinarian and human health-care professionals.

A direct benefit for patients will be the administration of antibiotics adapted to the identified offending bacterial pathogen. This will avoid blind prescription of antibiotics, an important determinant of the antimicrobial resistance issue at the global level and offer best chance to prevent severe complications in infected people and ultimately a fatal outcome.

Acknowledgements: We thank participants (see below) of a meeting held in May 2017 at the Institut Pasteur (Paris, France) for their contribution to the initial discussions on leptospirosis. CG and MP are members of the Global Leptospirosis Environmental Action Network (GLEAN), aimed to improve global and local strategies to prevent and control leptospirosis in high-risk populations. Participants: Koussay Dellagi, Mathieu Picardeau, Pascale Bourhy, Albert Ko, Cyrille Goarant, Serge Morand, Eric Bertherat, and Soledad Colombe.

REFERENCES

1. WHO. The "World malaria report 2019" at a glance 2019. Available from: https://www.who.int/news-room/feature-stories/detail/world-malaria-report-2019.
2. Bhatt S, Gething PW, Brady OJ, Messina JP, Farlow AW, Moyes CL, et al. The global distribution and burden of dengue. Nature. 2013;496:504-7.
3. Sigera PC, Amarasekara R, Rodrigo C, Rajapakse S, Weeratunga P, DeSilva NL, et al. Risk prediction for severe disease and better diagnostic accuracy in early dengue infection; the Colombo dengue study. BMC Infect Dis.

2019;19(680).
4. Utama IMS, Lukman N, Sukmawati DD, Alisjahbana B, Alam A, Murniati D, et al. Dengue viral infection in Indonesia: Epidemiology, diagnostic challenges, and mutations from an observational cohort study. PLoS Negl Trop Dis. 2019;13(e0007785).
5. Franco M, Mulder M, Gilman R, Smits H. Human brucellosis. Lancet Infect Dis. 2007;7:775-86.
6. McDermott JJ, Arimib SM. Brucellosis in sub-Saharan Africa: epidemiology, control and impact. Vet Microbiol/. 2002;90:111-34.
7. Crump JA, Morrissey AB, Nicholson WL, Massung RF, Stoddard RA, Galloway RL, et al. Etiology of severe non-malaria febrile illness in Northern Tanzania: a prospective cohort study. PLoS Negl Trop Dis. 2013;7:e2324.
8. Prasad N, Murdoch DR, Reyburn H, Crump JA. Etiology of Severe Febrile Illness in Low- and Middle-Income Countries: A Systematic Review. PLoS One. 2015;10:e0127962.
9. Mayxay M, Castonguay-Vanier J, Chansamouth V, Dubot-Pérès A, Paris DH, Phetsouvanh R, et al. Causes of non-malarial fever in Laos: a prospective study. Lancet Glob Health. 2013;1:e46-54.
10. Maude RR, Ghose A, Samad R, deJong HK, Fukushima M, Wijedoru L, et al. A prospective study of the importance of enteric fever as a cause of non-malarial febrile illness in patients admitted to Chittagong Medical College Hospital, Bangladesh. BMC Infect Dis 2016;16:567.
11. Chipwaza B, Mhamphi GG, Ngatunga SD, Selemani M, Amuri M, Mugasa JP, et al. Prevalence of bacterial febrile illnesses in children in Kilosa district, Tanzania. PLoS Negl Trop Dis 2015;9:e0003750.
12. Punjabi NH, Taylor WRJ, Murphy GS, Purwaningsih S, Picarima H, Sisson J, et al. Etiology of acute, non-malaria, febrile illnesses in Jayapura, northeastern Papua, Indonesia. Am J Trop Med Hyg 2012;86:46-51.
13. Halliday JEB, Carugati M, Snavely ME, Allan KJ, Beamesderfer J, Ladbury GAF, et al. Zoonotic causes of febrile illness in malaria endemic countries: a systematic review. Lancet Infect Dis. 2020;20:e27-e37.
14. Elven J, Dahal P, Ashley EA, Thomas NV, Shrestha P, Stepniewska K, et al. Non-malarial febrile illness: a systematic review of published aetiological studies and case reports from Africa, 1980-2015. BMC Med. 2020;18:279.
15. McDermott J, Grace D, Zinsstag J. Economics of brucellosis impact and control in low-income countries. Rev Sci Tech. 2013;32:249-61.
16. Sanhueza JM, G Baker MG, Benschop J, Collins-Emerson JM, Wilson PR, Heuer C. Estimation of the burden of leptospirosis in New Zealand. Zoonoses Public Health. 2020;67:167-76.
17. Mableson HE, Okello A, Picozzi K, Welburn SC. Neglected zoonotic diseases-the long and winding road to advocacy. PLoS Negl Trop Dis. 2014;8(e2800).
18. Goarant C, Picardeau M, Morand S, McIntyre KM. Leptospirosis under the bibliometrics radar: evidence for a vicious circle of neglect. J Glob Health 2019;9:010302.
19. Head MG, Brown RJ, Newell ML, Scott JAG, Batchelor J, Atun R. The allocation of USdollar;105 billion in global funding from G20 countries for infectious disease research between 2000 and 2017: a content analysis of investments. Lancet Glob Health. 2020;8(e1295-e1304).
20. FAO. The monetary impact of zoonotic diseases on society Evidence from three zoonoses in Kenya 2018. Available from: http://www.fao.org/3/i8968en/I8968EN.pdf.
21. Maze MJ, Biggs HM, Rubach MP, Galloway RL, Cash-Goldwasser S, Allan KJ, et al. Comparison of the Estimated Incidence of Acute Leptospirosis in the Kilimanjaro Region of Tanzania between 2007-08 and 2012-14. PLoS Negl Trop Dis 2016;10:e0005165.
22. Carugati M, Biggs HM, Maze MJ, Stoddard RA, Cash-Goldwasser S, Hertz JT, et al. Incidence of human brucellosis in the Kilimanjaro Region of Tanzania in the periods 2007-2008 and 2012-2014. Trans R Soc Trop Med Hyg. 2018;112:136-43.
23. Hartskeerl RA, Collares-Pereira M, Ellis WA. Emergence, control and re-emerging leptospirosis: dynamics of infection in the changing world. Clin Microbiol Infect. 2011;17:494-501.
24. Pijnacker R, Goris MG, Te Wierik MJ, Broens EM, van der Giessen JW, de Rosa M, et al. Marked increase in leptospirosis infections in humans and dogs in the Netherlands, 2014. Euro Surveill 2016;21.
25. Costa F, Hagan JE, Calcagno J, Kane M, Torgerson P, Martinez-Silveira MS, et al. Global Morbidity and Mortality of Leptospirosis: A Systematic Review. PLoS Negl Trop Dis 2015;9:e0003898.
26. Ellis WA. Animal Leptospirosis. In: Adler B, editor. Leptospira and Leptospirosis: Springer; 2014. p. 99-137.
27. Bierque E, Thibeaux R, Girault D, Soupé-Gilbert ME, Goarant C. A systematic review of Leptospira in water and soil environments. PLoS One. 2020;15(e0227055).
28. Goarant C. Leptospirosis: risk factors and management challenges in developing countries. Res Rep Trop Med. 2016;7:49-62.
29. Picardeau M, Bertherat E, Jancloes M, Skouloudis AN, Durski K, Hartskeerl RA. Rapid tests for diagnosis of leptospirosis: current tools and emerging technologies. Diagn Microbiol Infect Dis 2014;78:1-8.
30. Tubiana S, Mikulski M, Becam J, Lacassin F, Lefèvre P, Gourinat AC, et al. Risk factors and predictors of severe leptospirosis in New Caledonia. PLoS Negl Trop Dis 2013;7:e1991.
31. Adler B. Vaccines against leptospirosis. Curr Top Microbiol Immunol 2015;387:251-72.
32. Thornley CN, Baker MG, Weinstein P, Maas EW. Changing epidemiology of human leptospirosis in New Zealand. Epidemiol Infect 2002;129:29-36.
33. Limmathurotsakul D, Golding N, Dance DAB, Messina JP, Pigott DM, Moyes CL, et al. Predicted global distribution of Burkholderia pseudomallei and burden of melioidosis. Nature Microbiology. 2016;1:15008. doi: 10.1038/nmicrobiol.2015.8. http://www.nature.com/articles/nmicrobiol20158#supplementary-information.
34. Gassiep I, Armstrong M, Norton R. Human Melioidosis. Clinical microbiology reviews. 2020;33(2). Epub 2020/03/13. doi: 10.1128/cmr.00006-19.
35. Birnie E, Virk HS, Savelkoel J, Spijker R, Bertherat E, Dance DAB, et al. Global burden of melioidosis in 2015: a systematic review and data synthesis. Lancet Infect Dis. 2019;19:892-902.

36. Hin HS, Ramalingam R, Chunn KY, Ahmad N, Ab Rahman J, Mohamed MS. Fatal co-infection--melioidosis and leptospirosis. The American journal of tropical medicine and hygiene. 2012;87(4):737-40. Epub 2012/07/25. doi: 10.4269/ajtmh.2012.12-0165.
37. Sapian M, Khair MT, How SH, Rajalingam R, Sahhir K, Norazah A, et al. Outbreak of melioidosis and leptospirosis co-infection following a rescue operation. The Medical journal of Malaysia. 2012;67(3):293-7. Epub 2012/10/23.
38. Ngauy V, Lemeshev Y, Sadkowski L, Crawford G. Cutaneous melioidosis in a man who was taken as a prisoner of war by the Japanese during World War II. Journal of clinical microbiology. 2005;43(2):970-2. Epub 2005/02/08. doi: 10.1128/jcm.43.2.970-972.2005.
39. Suntornsut P, Wongsuwan N, Malasit M, Kitphati R, Michie S, Peacock SJ, et al. Barriers and Recommended Interventions to Prevent Melioidosis in Northeast Thailand: A Focus Group Study Using the Behaviour Change Wheel. PLoS Negl Trop Dis. 2016;10:e0004823.
40. Gibney KB, Cheng AC. Reducing the melioidosis burden: public health, chronic disease prevention, or improved case management? . Lancet Infect Dis. 2019;19:800-2.
41. Ismail N, McBride JW. Tick-Borne Emerging Infections: Ehrlichiosis and Anaplasmosis Clin Lab Med. 2017;37:317-40.
42. Elliott I, Pearson I, Dahal P, Thomas NV, Roberts T, Newton PN. Scrub typhus ecology: a systematic review of Orientia in vectors and hosts. Parasit Vectors. 2019;12(513).
43. Xu G, Walker DH, Jupiter D, Melby PC, Arcari CM. A review of the global epidemiology of scrub typhus. PLoS Negl Trop Dis. 2017;11(e0006062).
44. Bonell A, Lubell Y, Newton PN, Crump JA, Paris DH. Estimating the burden of scrub typhus: a systematic review. PLoS Negl Trop Dis. 2017;11(e0005838).
45. Yang L, Liang S, Wang X, Li X, Wu Y, Ma W. Burden of disease measured by disability-adjusted life years and a disease forecasting time series model of scrub typhus in Laiwu, China. PLoS Negl Trop Dis. 2015;9(e3420).
46. Ning C, Shuyi G, Tao Y, Hao Z, Zhang X. Epidemiological survey of human brucellosis in Inner Mongolia, China, 2010–2014: A high risk groups-based survey. J Infect Public Health. 2018;11:24-9.
47. Zhang N, Huang D, Wu W, Liu J, Liang F, Zhou B, et al. Animal brucellosis control or eradication programs worldwide: A systematic review of experiences and lessons learned Prev Vet Med. 2018;160:105-15.
48. Chappuis F, Alirol E, d'Acremont V, Bottieau E, Yansouni CP. Rapid diagnostic tests for non-malarial febrile illness in the tropics. Clinical Microbiology and Infection. 2013;19(5):422-31. doi: http://dx.doi.org/10.1111/1469-0691.12154.
49. Sea-Liang N, Sereemaspun A, Patarakul K, Gaywee J, Rodkvamtook W, Srisawat N, et al. Development of multiplex PCR for neglected infectious diseases. PLoS Negl Trop Dis 2019;13:e0007440.
50. Giry C, Roquebert B, Li-Pat-Yuen G, Gasque P, Jaffar-Bandjee MC. Simultaneous detection of chikungunya virus, dengue virus and human pathogenic Leptospira genomes using a multiplex TaqMan® assay. BMC Microbiol 2017;17:105.
51. Waggoner JJ, Abeynayake J, Balassiano I, Lefterova M, Sahoo MK, Liu Y, et al. Multiplex nucleic acid amplification test for diagnosis of dengue fever, malaria, and leptospirosis. J Clin Microbiol 2014;52:2011-8.
52. Abd El Wahed A, Weidmann M, Hufert FT. Diagnostics-in-a-Suitcase: Development of a portable and rapid assay for the detection of the emerging avian influenza A (H7N9) virus. J Clin Virol. 2015 Aug;69:16-21.
53. Weidmann M, Faye O, Faye O, AbdElWahed A, Patel P, Batejat C, et al. Development of Mobile Laboratory for Viral Hemorrhagic Fever Detection in Africa. J Infect Dis. 2018;218:1622-30.
54. Kotloff KL, Nataro JP, Blackwelder WC, Nasrin D, Farag TH, Panchalingam S, et al. Burden and aetiology of diarrhoeal disease in infants and young children in developing countries (the Global Enteric Multicenter Study, GEMS): a prospective, case-control study. Lancet. 2013;382:209-22.
55. Levine OS, O'Brien KL, Deloria-Knoll M, Murdoch DR, Feikin DR, DeLuca AN, et al. The Pneumonia Etiology Research for Child Health Project: a 21st century childhood pneumonia etiology study. Clin Infect Dis 2012;54.
56. Rhee C, Kharod GA, Schaad N, Furukawa NW, Vora NM, Blaney DD, et al. Global knowledge gaps in acute febrile illness etiologic investigations: A scoping review. PLoS Negl Trop Dis. 2019;13(e0007792).
57. AVMA. One health: A new professional imperative. 2008. Available from: https://www.avma.org/sites/default/files/resources/onehealth_final.pdf.
58. Robardet E, Bosnjak D, Englund L, Demetriou P, Martín PR, Cliquet F. Zero Endemic Cases of Wildlife Rabies (Classical Rabies Virus, RABV) in the European Union by 2020: An Achievable Goal. Tropical medicine and infectious disease. 2019;4(4). Epub 2019/10/03. doi: 10.3390/tropicalmed4040124.
59. Cárdenas L, Awada L, Tizzani P, Cáceres P, Casal J. Characterization and evolution of countries affected by bovine brucellosis (1996-2014). Transboundary and emerging diseases. 2019;66(3):1280-90. Epub 2019/02/11. doi: 10.1111/tbed.13144.
60. Currier RW, Widness JA. A Brief History of Milk Hygiene and Its Impact on Infant Mortality from 1875 to 1925 and Implications for Today: A Review. Journal of food protection. 2018;81(10):1713-22. Epub 2018/09/21. doi: 10.4315/0362-028x.jfp-18-186.
61. Dhiman RC. Emerging vector-borne zoonoses: eco-epidemiology and public health implications in India. Frontiers in public health. 2014;2:168. Epub 2014/10/18. doi: 10.3389/fpubh.2014.00168.
62. Zhang WY, Wang LY, Ding F, Hu WB, Soares Magalhaes RJ, Sun HL, et al. Scrub typhus in mainland China, 2006-2012: the need for targeted public health interventions. PLoS neglected tropical diseases. 2013;7(12):e2493. Epub 2014/01/05. doi: 10.1371/journal.pntd.0002493.
63. Thibeaux R, Girault D, Bierque E, Soupé-Gilbert ME, Rettinger A, Douyère A, et al. Biodiversity of Environmental Leptospira: Improving Identification and Revisiting the Diagnosis. Front Microbiol 2018;9:816.
64. Roger FL, Solano P, Bouyer J, Porphyre V, Berthier D, Peyre M, et al. Advocacy for identifying certain animal diseases as "neglected". PLoS Negl Trop Dis.

2017;11(e0005843).
65. Hopkins H, Bassat Q, Chandler C, Crump JA, Feasey NA, Ferrand RA, et al. Febrile Illness Evaluation in a Broad Range of Endemicities (FIEBRE): protocol for a multisite prospective observational study of the causes of fever in Africa and Asia. BMJ Open. 2020;10(e035632).
66. Pasteur Institut. Pasteur International Network 2020. Available from: https://www.pasteur.fr/en/international-en.
67. Pieracci EG, Hall AJ, Gharpure R, Haile A, Walelign E, Deressa A, et al. Prioritizing zoonotic diseases in Ethiopia using a one health approach. One Health. 2016;2:131-5.
68. Smith JK, Young MM, Wilson KL, Craig SB. Leptospirosis following a major flood in Central Queensland, Australia. Epidemiology and infection. 2013;141(3):585-90. Epub 2012/05/26. doi: 10.1017/S0950268812001021.
69. WHO. Control of Neglected Tropical Diseases. Available from: https://www.who.int/neglected_diseases/diseases/en/.
70. Murray CL, Barber RM, Foreman KJ, Ozgoren AA, Abd-Allah F, Abera SF, et al. Global, regional, and national disability-adjusted life years (DALYs) for 306 diseases and injuries and healthy life expectancy (HALE) for 188 countries, 1990-2013: quantifying the epidemiological transition. Lancet. 2015;386:2145-91.
71. Murray CJ, Ortblad KF, Guinovart C, Lim SS, Wolock TM, Roberts DA, et al. Global, regional, and national incidence and mortality for HIV, tuberculosis, and malaria during 1990-2013: a systematic analysis for the Global Burden of Disease Study 2013. Lancet. 2014;384:1005-70.
72. Torgerson PR, Hagan JE, Costa F, Calcagno J, Kane M, Martinez-Silveira MS, et al. Global Burden of Leptospirosis: Estimated in Terms of Disability Adjusted Life Years. PLoS Negl Trop Dis 2015;9:e0004122.