Eliminating visceral leishmaniasis in South Asia: the road ahead

Suman Rijal and colleagues highlight lessons from a regional collaboration to eliminate visceral leishmaniasis and identify priorities for the post-elimination plan

Devastating epidemics of visceral leishmaniasis, also known as kala-azar, have been recorded on the Indian subcontinent since the early 19th century,[1] most commonly affecting poor people.[2] The three most affected countries in South Asia are India, Bangladesh, and Nepal. Sporadic cases have been reported in Bhutan and Sri Lanka. Box 1 describes key features of kala-azar in South Asia. Efforts to control the disease have had limited impact.[3] Until recently, these countries accounted for more than 50% of the global disease burden.[4] Sustained elimination efforts have led to a steady decline in recent years. However, some transmission continues and outbreaks in non-immune populations remain likely. As the number of kala-azar cases becomes negligible, newer tools and strategies will be required for diagnosis, treatment, and vector control.

Regional collaboration to eliminate kala-azar

In 2005, the governments of three endemic countries in South Asia—Bangladesh, India, and Nepal—jointly established a regional alliance to eliminate kala-azar, supported by the World Health Organization.[5 6] The kala-azar elimination programme is the only regional collaboration globally to tackle this disease. It set a target to decrease the incidence of kala-azar to a level at which it was no longer a public health problem by 2015. That deadline has now been extended to 2020.[7] The development of the oral drug miltefosine[8] and a rapid diagnostic test based on the rK39 antigen[9] have had a critical role in early diagnosis and providing effective treatment to reduce the disease burden. Additionally, the programme has focused on vector control and improved surveillance to reduce transmission and improve case detection. Figure 1 depicts the strategy with the key outcomes in this initiative to achieve the target of less than one per 10 000 population.

The number of kala-azar cases in these countries has declined steadily from over 77 000 reported cases in 1992 to fewer than 7000 cases in 2016 (fig 2). In 2016, 242 new cases were reported in Nepal, 255 in Bangladesh, and 6249 in India.[10] Nepal achieved the elimination threshold in 2013,[11] and Bangladesh in 2016.[12] In India, some more effort will be required, as 8% of endemic units were still above the threshold at the end of 2017.

The impact of the kala-azar elimination programme has not been systematically evaluated, but it is likely that sustained focus and collaborative efforts through the programme have contributed to the declining incidence. Moreover, the level of reporting has improved, providing a more accurate estimate of the disease burden. Possibly, the free treatment offered by public health services under the programme has led to better notification of the disease. In India, for example, underreporting declined to a factor of 1.2 in 2015 from a factor of three to eight observed in 2003 and 2005.[13-16]

Further challenges

Caution is needed as a resurgence of kala-azar is possible. The programme did not target "elimination of the pathogen" and thus some transmission continues. Figure 2 shows that the number of cases in India seems to follow roughly 15 year cycles. This makes it difficult to assess the effect of interventions to control the disease, as the downward trend may be the result of the "natural" fluctuation of the disease.[17] Communities that had some herd immunity in the past may gradually be becoming fully susceptible.[18] The rising trend in post-kala-azar dermal leishmaniasis[19] together with the emergence of coinfection with HIV is concerning.[20] Patients with these conditions may serve as reservoirs of infection, perpetuating transmission even when the elimination targets are reached.[20 21] Treatments for both conditions are far from ideal.[19 20]

Priorities to sustain elimination

In the post-elimination phase, surveillance needs to be maintained while detection and control strategies will need to be modified.

Improved diagnostic tools

The current rapid diagnostic test detects antibodies against rK39 antigen. To confirm

KEY MESSAGES

- The kala-azar elimination programme has made substantial progress in reducing incidence in India, Bangladesh, and Nepal
- With increasing incidence of post-kala-azar dermal leishmaniasis and HIV-*Leishmania* coinfection low grade transmission continues and there is a risk of outbreaks
- Investing in the development of new drugs and diagnostics as well as innovative vector control and surveillance strategies is crucial to sustain the progress in elimination

Box 1: Key features of kala-azar in Bangladesh, India, and Nepal

- Visceral leishmaniasis or kala-azar is caused by *Leishmania donovani* parasites and transmitted by the sand fly, *Phlebotomus argentipes*. Humans are considered the only reservoirs of infection
- After an incubation period of 2-6 months, patients develop a syndrome characterised by fever, splenomegaly, wasting, and anaemia. It is fatal if left untreated
- Demonstration of parasites in a smear or culture of aspirate from spleen, bone marrow, or lymph node is required to confirm the diagnosis. Alternatively, serological evidence in a patient with recent onset of febrile splenomegaly in endemic areas will suffice
- Treatment regimens vary by region. In Asia, a single dose infusion of liposomal amphotericin B is the first treatment, with several combination regimens as alternatives
- Around 5-10% of patients develop post-kala-azar dermal leishmaniasis 6 months or more after the disease has apparently been cured. They are a potential source of infection

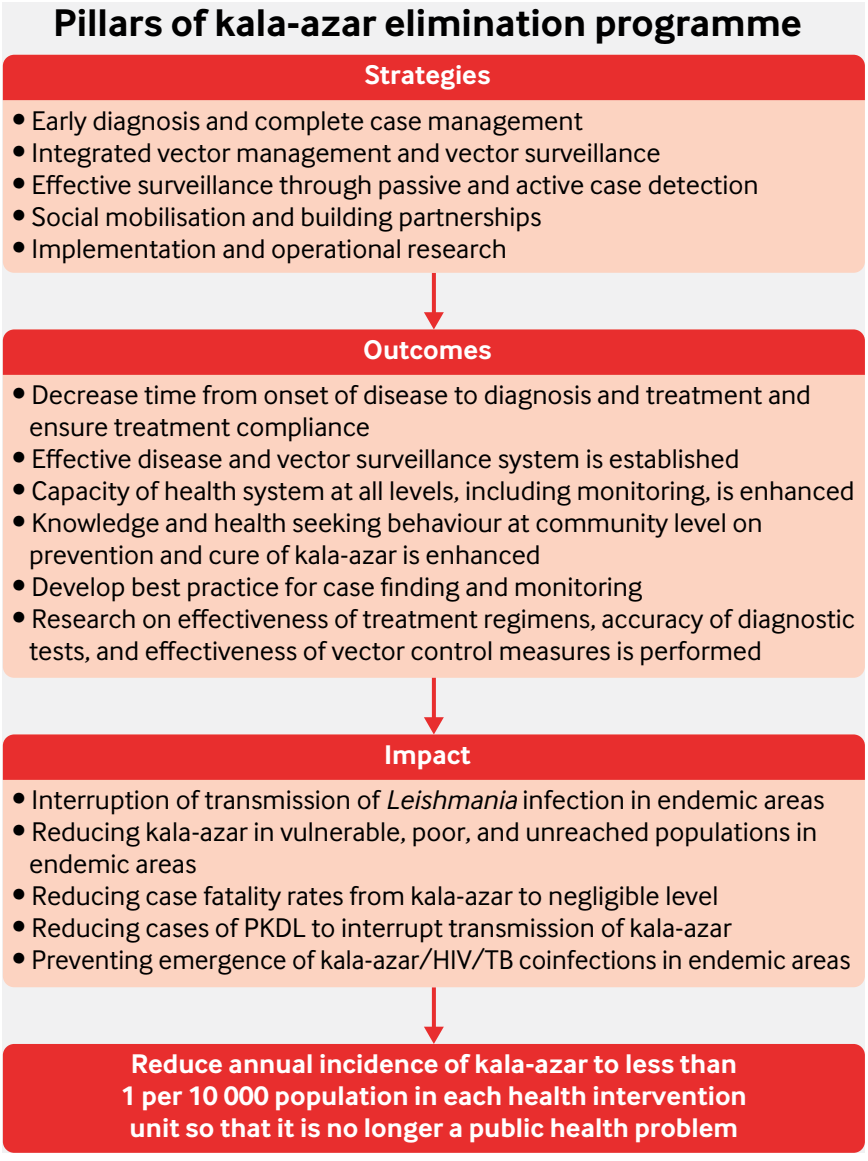

Fig 1 | Strategy for the kala-azar elimination programme in India, Nepal, and Bangladesh (adapted from WHO regional strategic framework for elimination of kala-azar[42]). PKDL=post-kala-azar dermal leishmaniasis

diagnosis and start treatment a positive result must be interpreted in conjunction with clinical features—that is, fever for two weeks and a palpable spleen. On its own, the test is not specific for the acute stage of the disease, and is also positive in latent carriers and in cured patients. The combination with a clinical case definition induces a delay of two weeks before the patient is diagnosed. Decreasing the time between onset of symptoms and diagnosis might help reduce transmission.[22]

The kala-azar elimination programme has benefited greatly from this diagnostic test for detection of cases. However, the test may become inadequate in the post-elimination phase as its positive predictive value may decrease rapidly when near elimination is achieved. Many patients with a false positive result risk being given treatment for kala-azar while the actual cause of their persistent fever (brucellosis, rickettsiosis, tuberculosis, etc) is not dealt with. A more specific test will be required, preferably based on antigen detection.[23 24] Table 1 lists some diagnostics test under development that might overcome the limitations of the current test and be more appropriate in the post-elimination era.

Newer drugs

The current drug regimens, while allowing progress towards eliminating kala-azar, will probably be inadequate for the post-elimination phase.[25] Based on WHO recommendations, the kala-azar elimination programme replaced miltefosine with a single dose infusion of liposomal amphotericin B (AmBisome) as first line treatment in 2013. AmBisome has shown greater efficacy and improved compliance, but it requires a strict cold chain. AmBisome has been used successfully in the attack phase of the programme in India. However, the entire programme (ie, primary kala-azar, relapses, post-kala-azar dermal leishmaniasis, and HIV-kala-azar cases) is now reliant on a single medicine produced by a single manufacturer. Relapses have been observed with this treatment.[25]

Paromomycin-miltefosine combination therapy is recommended as an alternative where a cold chain cannot be ensured. This regimen includes 10 days of injections with paromomycin. Miltefosine is potentially teratogenic, which limits its use in women. Current trials in India and Bangladesh (CTRI/2017/04/008421, CTRI/2015/05/005807) aim to evaluate the efficacy of different regimens of the AmBisome-miltefosine combination to reduce treatment duration, relapses, and toxicity in patients with post-kala-azar dermal leishmaniasis and HIV coinfection.

Most of these trials are using repurposed drugs developed for other indications and not according to a target product profile reflecting the requirements of a sustainable

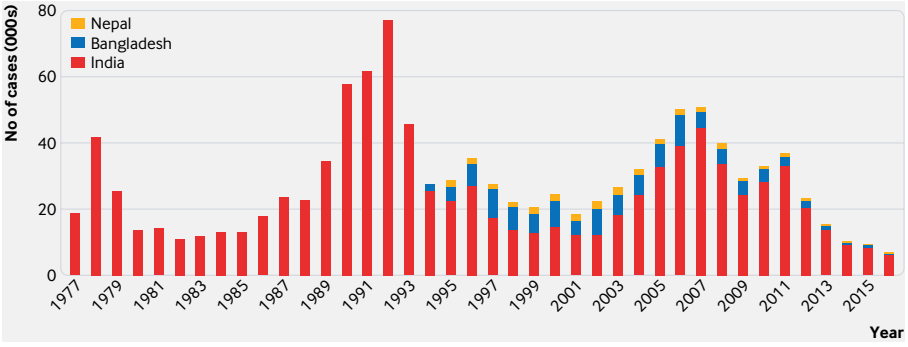

Fig 2 | Number of kala-azar cases reported by Nepal, Bangladesh, and India, 1977-2016 (WHO/Global Health Repository and country data[10])

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Table 1 | Diagnostic tests for kala-azar and post-kala-azar dermal leishmaniasis in development

Test	Description
Antibody detection rapid diagnostic test (RDT)	
Lateral flow immunochromatographic RDT: leishmaniasis IgG1 RDT[43] (Coris Bioconcept, Belgium)	Rapid diagnostic test to detect anti-*Leishmania* IgG1 as a potential biomarker of post-chemotherapeutic relapse. Raised levels of specific IgG1 were associated with treatment failure and relapse, whereas no or low IgG1 levels were detected in patients whose visceral leishmaniasis had been cured. Further evaluation is needed to determine its usefulness in the field (AfriKADIA consortium expected to yield results in 2020)
Antigen detection	
Urine—ELISA[44]: *Leishmania* antigen detection ELISA (InBios International, Seattle, USA) *Leishmania* antigen ELISA (visceral leishmaniasis ELISA) (Kalon Biologicals, UK)	Non-invasive test to detect urinary *Leishmania* antigens during the acute stage and monitor their clearance when a cure is achieved. Evaluated in Asia and Africa with good sensitivity (>90%) and specificity. Further refinement of the test is needed using more samples from endemic regions to define their usefulness in monitoring treatment. Could replace the invasive splenic aspirations and serve as a standardised tool to measure the effectiveness of emerging treatment regimens
Urine—agglutination: KAtex latex agglutination test[45] (Kalon Biologicals, UK)	Urinary *Leishmania* antigen detection agglutination test. Evaluated in Asia, Africa, Europe, and Latin America. Low sensitivity, though specificity good. Potential for evaluating a cure
Nucleic acid amplification tests	
Blood—loop-mediated isothermal amplification (LAMP)[46]: Loopamp *Leishmania* detection kit (Eiken Chemical, Japan)	Loopamp is the first LAMP test available as a kit which has been validated for kala-azar and commercially available. It is rapid, simple, and highly specific. Diagnosis of kala-azar using peripheral blood in Asia and Africa showed high sensitivity (>90%) and excellent specificity, with >90% sensitivity and specificity in diagnosis of post-kala-azar dermal leishmaniasis. Needs further validation as a test for cure
Recombinase polymerase amplification (RPA) assay[47]: *Leishmania donovani* RPA assay	Field based test for diagnosis in areas with low resources. Feasibility was shown to be good. Further validation needed at more sites

elimination programme. Ideally, a new drug should be able to be taken orally and combine high efficacy with an excellent safety profile for deployment in remote areas with poor health infrastructures. Half of all patients are children, so drug development should take this into account. An optimal drug combination would have a short (<10 days) treatment duration, different mechanisms of action to offer protection from resistance, a good safety profile, and no interaction with other drugs commonly used in these areas, such as antimalarials.

Several pharmaceutical research groups have invested heavily in discovering a drug targeting *Leishmania* parasites. Six new chemically diverse drugs, targeting five different molecular mechanisms, are in the late stages of development (table 2). All of these are oral drugs and reduce the parasite load by >95% in animal models of kala-azar when given for up to 10 days.[25] Given the typical attrition rates in the drug discovery process, one or two compounds could be registered by 2025, providing a completely different treatment for the post-elimination phase. Strategies must be developed to ensure compliance with these new treatments, similar to the directly observed treatment short course used for tuberculosis, to prevent emergence of resistance. Monitoring the safety and effectiveness of these treatments and emergence of drug resistance will be required.

Vector control measures to reduce transmission

Measures to control vectors have primarily been indoor residual spraying of insecticides in endemic villages reporting kala-azar cases in the preceding three years. A toolkit for monitoring and evaluation of entomological interventions was developed within the kala-azar elimination programme.[27] Pyrethroid spraying has been shown to be effective in reducing sand flies in carefully controlled experiments.[28] However, field studies suggest that the level of vectors has not declined significantly in villages treated by indoor residual spraying.[29] Spraying also requires a lot of equipment, is expensive, and is often not easily acceptable to communities, making it unsustainable in the long term.

In view of these drawbacks, researchers looked for alternatives that were cost effective, had a longer period of efficacy, and were easy to use and sustain. Trials in Bangladesh, India, and Nepal have shown reduction of sand fly density using reimpregnated commercial bed nets and longlasting insecticide treated bed nets.[30 31] However, they have not been shown to confer protection against visceral leishmaniasis in a cluster randomised trial in India and Nepal.[32] A comparative study in endemic villages in Bangladesh showed a greater decrease in the incidence of visceral leishmaniasis in one area where people slept under bed nets impregnated with a slow release insecticide, KO Tab, compared with the control area.[33] Durable wall lining has also shown promise in controlling sand fly density, although the initial cost is high.[34] Other tools being evaluated include wall paint containing three insecticides, including a larvicide, and an insecticide repellent combination for canine leishmaniasis.[35]

We must complete our knowledge of vector bionomics and behaviour to allow for better designed and more effective tools for vector control.[36]

Table 2 | Preclinical and clinical drug candidates in the late stages of development for visceral leishmaniasis (adapted from Mowbray[26])

Drug	Class	Mode of action
DNDi-0690	Nitroimidazole	Bioactivation by parasitic nitroreductase NTR2
DNDi-6148	Oxaborole	Unknown but active against *Leishmania* strains
XE408	Proteasome inhibitor	Inhibition of parasitic proteasome
GSK3494245/1305143	Proteasome inhibitor	Inhibition of parasitic proteasome
GSK-3186899/DDD853651	Pyrazolopyrimidine	Inhibition of *Leishmania* CRK12 kinase
DNDi-5561	Aminopyrazole	Unknown

Case detection and epidemiological surveillance

Epidemiological surveillance of kala-azar has improved considerably under the kala-azar elimination programme, and underreporting is now minimal. As the incidence of the disease declines, awareness and knowledge of the disease will probably fall in both patients and clinicians. Active screening for case detection will stop. To be effective and sustainable in the post-elimination era, systems for case detection, notification, and surveillance will need to be redesigned.

People affected by kala-azar continue to present at a late stage to primary health centres, and diagnosis is often delayed.[22 37 38] Furthermore, these primary health centres are poorly resourced, making it difficult to provide good quality

care. Kala-azar control programmes are largely organised vertically and are often disconnected from the reality in the field. At primary healthcare level, physicians, nurses, and other medical professionals deal with patients presenting with a wide spectrum of complaints.[39] An integrated approach is required for the surveillance and management of fever at primary care level in kala-azar endemic settings to ensure that no cases are missed. A syndrome based approach to clinical management of fever must be adopted. This would include kala-azar as a differential diagnosis and allow systematic testing with a rapid diagnostic test for kala-azar and other conditions in patients with fever for more than two weeks.[40] Cooperation with the private health sector will be crucial to ensure that all patients are reached.

New tools and strategies are required for epidemiological surveillance in the post-elimination phase. These include tools for detection of kala-azar and post-kala-azar dermal leishmaniasis when their incidence is very low, an adequate response strategy to an outbreak, and proxy markers for sand fly infectivity.[11] Population based serosurveillance is a powerful tool to examine trends in infection rates and gauge the effect of the kala-azar elimination programme. Established health and demographic surveillance systems[41] in the region can contribute by monitoring trends in kala-azar incidence and seroprevalence over a longer time.

Maintaining success

Continued vigilance will be required to sustain the gains achieved through kala-azar elimination efforts. The programme will need to evolve and realign strategies to meet the requirements of this post-elimination phase. This will necessitate proportionate investments in research and development of new tools, training of health workers, and logistics and infrastructure to improve the quality of primary care. Commitment to eliminating the scourge of kala-azar from this region and globally must continue.

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1 Peters W, Prasad LSN. Kala-azar in India—its importance as an issue in public health. In: *Proceedings of the Indo-UK Workshop on Leishmaniasis*. New Delhi, India. Indian Council of Medical Research, 1983.
2 Boelaert M, Meheus F, Sanchez A, et al. The poorest of the poor: a poverty appraisal of households affected by visceral leishmaniasis in Bihar, India. *Trop Med Int Health* 2009;14:639-44. doi:10.1111/j.1365-3156.2009.02279.x
3 Bora D. Epidemiology of visceral leishmaniasis in India. *Natl Med J India* 1999;12:62-8.
4 Alvar J, Vélez ID, Bern C, et al, WHO Leishmaniasis Control Team. Leishmaniasis worldwide and global estimates of its incidence. *PLoS One* 2012;7:e35671. doi:10.1371/journal.pone.0035671
5 World Health Organization Regional Office for South-East Asia. Regional strategic framework for elimination of kala-azar from the South-East Asia region (2005-2015). 2005. http://apps.searo.who.int/pds_docs/B0211.pdf
6 Hirve S, Kroeger A, Matlashewski G, et al. Towards elimination of visceral leishmaniasis in the Indian subcontinent-translating research to practice to public health. *PLoS Negl Trop Dis* 2017;11:e0005889. doi:10.1371/journal.pntd.0005889
7 WHO. Accelerating work to overcome the global impact of neglected tropical diseases - a roadmap for implementation. 2012. https://www.who.int/neglected_diseases/NTD_RoadMap_2012_Fullversion.pdf?ua=1
8 Sundar S, Jha TK, Thakur CP, et al. Oral miltefosine for Indian visceral leishmaniasis. *N Engl J Med* 2002;347:1739-46. doi:10.1056/NEJMoa021556
9 Boelaert M, Verdonck K, Menten J, et al. Rapid tests for the diagnosis of visceral leishmaniasis in patients with suspected disease. *Cochrane Database Syst Rev* 2014;6:CD009135. doi:10.1002/14651858.CD009135.pub2.
10 WHO. Global Health Observatory Data Repository (last updated 15 Sep 2017). http://apps.who.int/gho/data/node.main.NTDLEISHVNUM?lang=en
11 Olliaro PL, Shamsuzzaman TA, Marasini B, et al. Investments in research and surveillance are needed to go beyond elimination and stop transmission of leishmania in the Indian subcontinent. *PLoS Negl Trop Dis* 2017;11:e0005190. doi:10.1371/journal.pntd.0005190
12 Mondal D. Elimination efforts in Bangladesh: lessons learned and challenges remaining. SPEAK India Delhi, India, 3-5 November 2016. https://speakindia.org.in/presentations
13 Singh SP, Reddy DC, Rai M, Sundar S. Serious underreporting of visceral leishmaniasis through passive case reporting in Bihar, India. *Trop Med Int Health* 2006;11:899-905. doi:10.1111/j.1365-3156.2006.01647.x
14 Singh VP, Ranjan A, Topno RK, et al. Estimation of under-reporting of visceral leishmaniasis cases in Bihar, India. *Am J Trop Med Hyg* 2010;82:9-11. doi:10.4269/ajtmh.2010.09-0235
15 Mubayi A, Castillo-Chavez C, Chowell G, et al. Transmission dynamics and underreporting of Kala-azar in the Indian state of Bihar. *J Theor Biol* 2010;262:177-85. doi:10.1016/j.jtbi.2009.09.012
16 Sridhar S. Learnings for VL surveillance and monitoring systems. SPEAK India, Delhi, India. 24-25 April 2017. https://speakindia.org.in/presentations.
17 Dye C, Wolpert DM. Earthquakes, influenza and cycles of Indian kala-azar. *Trans R Soc Trop Med Hyg* 1988;82:843-50. doi:10.1016/0035-9203(88)90013-2
18 Le Rutte EA, Chapman LAC, Coffeng LE, et al. Policy recommendations from transmission modeling for the elimination of visceral leishmaniasis in the Indian subcontinent. *Clin Infect Dis* 2018;66(suppl_4):S301-8. doi:10.1093/cid/ciy007
19 Zijlstra EE, Alves F, Rijal S, Arana B, Alvar J. Post-kala-azar dermal leishmaniasis in the Indian subcontinent: a threat to the South-East Asia Region Kala-azar Elimination Programme. *PLoS Negl Trop Dis* 2017;11:e0005877. doi:10.1371/journal.pntd.0005877
20 Akuffo H, Costa C, van Griensven J, Burza S, Moreno J, Herrero M. New insights into leishmaniasis in the immunosuppressed. *PLoS Negl Trop Dis* 2018;12:e0006375. doi:10.1371/journal.pntd.0006375
21 Mondal D, Bern C, Ghosh D, et al. Quantifying the infectiousness of post-kala-azar dermal leishmaniasis towards sandflies. *Clin Infect Dis* 2018. [Epub ahead of print]. doi:10.1093/cid/ciy891
22 Medley GF, Hollingsworth TD, Olliaro PL, Adams ER. Health-seeking behaviour, diagnostics and transmission dynamics in the control of visceral leishmaniasis in the Indian subcontinent. *Nature* 2015;528:S102-8. doi:10.1038/nature16042
23 Chappuis F, Sundar S, Hailu A, et al. Visceral leishmaniasis: what are the needs for diagnosis, treatment and control?*Nat Rev Microbiol* 2007;5:873-82. doi:10.1038/nrmicro1748
24 Peeling RW, Mabey D. Diagnostics for the control and elimination of neglected tropical diseases. *Parasitology* 2014;141:1789-94. doi:10.1017/S0031182014000973
25 Alves F, Bilbe G, Blesson S, et al. Recent development of visceral leishmaniasis treatments: *success, pitfalls,*

*and perspectives. Microbiol Rev*2018;31:e00048-18. doi:10.1128/CMR.00048-18
26 Mowbray CE. Antileishmanial drug discovery: past, present and future perspectives. In: Rivas L, Gil C, eds. *Drug discovery for leishmaniasis*. Royal Society of Chemistry, 2018:24-36.
27 Special Programme for Research and Training in Tropical Diseases (WHO/TDR). Monitoring and evaluation toolkit for indoor residual spraying: kala-azar elimination in Bangladesh, India and Nepal. 2010. https://www.who.int/tdr/publications/tdr-research-publications/irs_toolkit/en/
28 Coleman M, Foster GM, Deb R, et al. DDT-based indoor residual spraying suboptimal for visceral leishmaniasis elimination in India. *Proc Natl Acad Sci U S A* 2015;112:8573-8. doi:10.1073/pnas.1507782112
29 Poché DM, Garlapati RB, Mukherjee S, et al. Bionomics of Phlebotomus argentipes in villages in Bihar, India with insights into efficacy of IRS-based control measures. *PLoS Negl Trop Dis* 2018;12:e0006168. doi:10.1371/journal.pntd.0006168
30 Picado A, Das ML, Kumar V, et al. Effect of village-wide use of long-lasting insecticidal nets on visceral leishmaniasis vectors in India and Nepal: a cluster randomized trial. *PLoS Negl Trop Dis* 2010;4:e587. doi:10.1371/journal.pntd.0000587
31 Chowdhury R, Faria S, Huda MM, et al. Control of Phlebotomus argentipes (Diptera: Psychodidae) sand fly in Bangladesh: a cluster randomized controlled trial. *PLoS Negl Trop Dis* 2017;11:e0005890. doi:10.1371/journal.pntd.0005890
32 Picado A, Singh SP, Rijal S, et al. Longlasting insecticidal nets for prevention of Leishmania donovani infection in India and Nepal: paired cluster randomised trial. *BMJ* 2010;341:c6760. doi:10.1136/bmj.c6760
33 Mondal D, Huda MM, Karmoker MK, et al. Reducing visceral leishmaniasis by insecticide impregnation of bed-nets, Bangladesh. *Emerg Infect Dis* 2013;19:1131-4. doi:10.3201/eid1907.120932
34 Huda MM, Kumar V, Das ML, et al. Entomological efficacy of durable wall lining with reduced wall surface coverage for strengthening visceral leishmaniasis vector control in Bangladesh, India and Nepal. *BMC Infect Dis* 2016;16:539. doi:10.1186/s12879-016-1881-8
35 Dumont P, Fankhauser B, Bouhsira E, et al. Repellent and insecticidal efficacy of a new combination of fipronil and permethrin against the main vector of canine leishmaniosis in Europe (Phlebotomus perniciosus). *Parasit Vectors* 2015;8:49. doi:10.1186/s13071-015-0683-y
36 Cameron MM, Acosta-Serrano A, Bern C, et al. Understanding the transmission dynamics of Leishmania donovani to provide robust evidence for interventions to eliminate visceral leishmaniasis in Bihar, India. *Parasit Vectors* 2016;9:25. doi:10.1186/s13071-016-1309-8
37 Hirve S, Boelaert M, Matlashewski G, et al. Transmission dynamics of visceral leishmaniasis in the indian subcontinent—a systematic literature review. *PLoS Negl Trop Dis* 2016;10:e0004896. doi:10.1371/journal.pntd.0004896
38 Hasker E, Singh SP, Malaviya P, et al. Management of visceral leishmaniasis in rural primary health care services in Bihar, India. *Trop Med Int Health* 2010;15(Suppl 2):55-62. doi:10.1111/j.1365-3156.2010.02562.x
39 Singh OP, Hasker E, Boelaert M, Sundar S. Elimination of visceral leishmaniasis on the Indian subcontinent. *Lancet Infect Dis* 2016;16:e304-9. doi:10.1016/S1473-3099(16)30140-2
40 Hercik C, Cosmas L, Mogeni OD, et al. A combined syndromic approach to examine viral, bacterial, and parasitic agents among febrile patients: a pilot study in Kilombero, Tanzania. *Am J Trop Med Hyg* 2018;98:625-32. doi:10.4269/ajtmh.17-0421
41 Malaviya P, Picado A, Hasker E. Health & demographic surveillance system profile: the Muzaffarpur-TMRC Health and Demographic Surveillance System. *Int J Epidemiol* 2014;43:1450-7. doi:10.1093/ije/dyu178
42 WHO. Regional strategic framework for elimination of kala-azar from the South-East Asia Region (2011-2015). http://www.who.int/iris/handle/10665/205826
43 Bhattacharyya T, Ayandeh A, Falconar AK, et al. IgG1 as a potential biomarker of post-chemotherapeutic relapse in visceral leishmaniasis, and adaptation to a rapid diagnostic test. *PLoS Negl Trop Dis* 2014;8:e3273. doi:10.1371/journal.pntd.0003273
44 Vallur AC, Tutterrow YL, Mohamath R, et al. Development and comparative evaluation of two antigen detection tests for visceral leishmaniasis. *BMC Infect Dis* 2015;15:384. doi:10.1186/s12879-015-1125-3
45 Boelaert M, El-Safi S, Hailu A, et al. Diagnostic tests for kala-azar: a multi-centre study of the freeze-dried DAT, rK39 strip test and KAtex in East Africa and the Indian subcontinent. *Trans R Soc Trop Med Hyg* 2008;102:32-40. doi:10.1016/j.trstmh.2007.09.003
46 Adams ER, Schoone G, Versteeg I, et al. Development and evaluation of a novel loop-mediated isothermal amplification assay for diagnosis of cutaneous and visceral leishmaniasis. *J Clin Microbiol* 2018;56:p.ii:e00386-18. doi:10.1128/JCM.00386-18
47 Mondal D, Ghosh P, Khan MA, et al. Mobile suitcase laboratory for rapid detection of Leishmania donovani using recombinase polymerase amplification assay. *Parasit Vectors* 2016;9:281. doi:10.1186/s13071-016-1572-8

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