

Review

The antipoverty vaccines

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Abstract

The neglected tropical diseases represent a group of parasitic and bacterial diseases, occurring primarily in rural areas or impoverished urban areas of developing countries. Because of their chronic and stigmatizing character and their impact on child development, pregnancy outcomes, and worker productivity, the neglected tropical diseases are considered poverty-promoting conditions. Through the activities of public–private partnerships, first or second-generation recombinant vaccines for three of these conditions—hookworm, leishmaniasis, and schistosomiasis, have undergone early development and clinical testing. However, through the acquisition of extensive bioinformatics information or animal model testing for several other neglected tropical diseases pathogens, it is possible to consider new generation vaccines as well for amebiasis, Buruli ulcer, Chagas disease, Chlamydia infections (including trachoma), leprosy, leptospirosis, and the treponematoses. Early development of such antipoverty vaccines will require the establishment of product development public–private partnerships and partnerships with innovative developing countries where these diseases are endemic.

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1. Introduction

The 21st century began with new views about the importance of tackling health as a means to poverty reduction. In January 2000, then World Health Organization Director General, Dr. Gro Harlem Brundtland, launched the Commission on Macroeconomics and Health [1]. The Commission was charged with analyzing the impact of health on development and exploring ways in which investments in health would translate into economic growth in developing countries. The resulting 2001 report confirmed a profound relationship between disease and chronic poverty, while subsequent papers identified a number of complex mechanisms underlying the association [2–4]. A second landmark event also occurred in 2000, when 147 heads of state met at the United Nations to adopt eight Millennium Development Goals (MDGs) for achieving poverty reduction through sustainable development [5,6].

The sixth MDG (MDG 6) focuses on infectious diseases, with a specific emphasis on reducing the incidence of HIV/AIDS, malaria, and “other diseases”. An important component of these other diseases is the neglected tropical diseases—a group of 13 poverty-promoting parasitic and bacterial infections, which include the soil-transmitted helminth infections, schistosomiasis, lymphatic filariasis, onchocerciasis, and trachoma. Together, HIV/AIDS, malaria and the neglected tropical diseases account for more than 4.5 million annual deaths. More important than the deaths caused by the neglected tropical diseases are the chronic and disabling features of this group of diseases. Using a metric known as the DALY (disability-adjusted life year, i.e., the numbers of life years lost from premature death or disability), HIV/AIDS and malaria result in approximately 84.5 and 46.5 million DALYs lost annually, respectively [7], while the neglected tropical diseases cause an estimated 56.6 million DALYs annually [8]. Along with lower respiratory tract infections and diarrheal diseases, HIV/AIDS, malaria, tuberculosis and the neglected tropical diseases constitute the most important communicable diseases of humankind [8].

In Sub-Saharan Africa and other parts of the developing world, HIV/AIDS, malaria, and the neglected tropical dis-

eases have a special link to poverty because of their impact on childhood development and education, maternal health and perinatal outcomes, and worker productivity. Through such mechanisms, these infections not only occur in the setting of poverty but, in addition, they also promote poverty [9]. Therefore, the successful development of new vaccines to improve immunity to these conditions represents a potentially promising antipoverty intervention. This review identifies several of the major neglected conditions identified as causes of poverty in developing countries and the role of antipoverty vaccines in achieving MDG 6. Because of the extensive literature on vaccines for HIV/AIDS, malaria, and tuberculosis (reviewed in refs. [10–12]), we focus here on efforts to develop vaccines for the lesser known “other diseases” including the neglected tropical diseases (e.g., hookworm, schistosomiasis, Chagas disease, leishmaniasis, amebiasis, trachoma and genital Chlamydia infections, Buruli ulcer, leprosy, leptospirosis, and syphilis) and drug addiction vaccines. Such vaccines would be considered orphan products with either small or non-existent commercial markets. The vaccines discussed here are in early development, e.g., at the preclinical testing stage or in Phase 1 or 2 clinical trials, and will require innovative mechanisms for their final commercial development and global access.

2. The poverty-promoting conditions

The neglected tropical diseases refer to a set of poverty-promoting conditions that afflict the rural poor in low-income countries, as well as some impoverished urban dwellers living in the slums of large cities in developing countries [9]. The core group of 13 neglected tropical diseases includes 7 helminth infections—ascariasis, trichuriasis, hookworm, lymphatic filariasis (LF), onchocerciasis, dracunculiasis, schistosomiasis; 3 protozoan infections—Chagas disease, human African trypanosomiasis, and leishmaniasis; and 3 bacterial infections—trachoma, leprosy, and Buruli ulcer [9,13,14]. These are ancient conditions that have plagued humankind for centuries, and are sometimes also referred to as the “biblical diseases” [14]. Because of their link to poverty and their impact on reducing worker productivity,

the neglected tropical diseases have a stigma attached to them, and this stigma is exacerbated when these diseases cause disfigurement such as in the cases of LF, onchocerciasis, leprosy, and Buruli ulcer [9,14]. Some clinicians and public health workers supplement this list with additional helminth infections (e.g., cysticercosis, echinococcosis, and food borne trematode infections), bacterial infections (e.g., leptospirosis, syphilis and other treponematoses), protozoan infection, amebiasis, and selected viral infections (e.g., rabies and the flavivirus infections, dengue, Japanese encephalitis, and yellow fever).

A list of the major poverty-promoting conditions in developing countries for which vaccine research and development programs have been initiated is shown in Table 1. These conditions were selected on the basis of their debilitating features, and their ability to impair child growth and intellectual development, to promote maternal and perinatal mortality during pregnancy, and/or to affect worker productivity [14]. Moreover, many of the diseases listed here are chronic in nature, so that their impact is frequently felt throughout both childhood and adulthood. Together the vaccines for these poverty-promoting conditions could prevent 207 million DALYs annually, approximately 60% of the global disability resulting from infectious diseases [7,8]. Discussed below are mechanisms by which the major disease conditions listed in Table 1 contribute to poverty, and how new programs to promote orphan vaccine development could significantly promote poverty reduction.

3. Impact on child health and development

An estimated 2 billion people are infected with three major soil-transmitted helminths, *Ascaris lumbricoides*, *Trichuris trichiura*, and the hookworms, and the two major schistosomes, *Schistosoma haematobium* and *Schistosoma mansoni* [15]. Together, these helminth infections result in approximately 415,000 annual deaths and 43.5 million DALYs [8]. School-aged children harbor higher worm burdens than any other group [16], and much of this helminth-associated disability results from the profound impact of chronic moderate and heavy infections on childhood nutrition (especially anemia), physical growth and fitness (reviewed in refs. [8,13,14,17,18]). Equally important, chronic polyparasitism results in deficits in working memory and other neurocognitive functions (reviewed in refs. [8,19]), with specific effects attributable to hookworm [20], trichuriasis [21], and schistosomiasis [22]. Together these effects translate into reduced school performance and attendance [19,23] probably accounting for the observation that chronic hookworm infection in childhood reduces future adult earnings [24].

The relationship between helminth infections and physical and mental deficits in children observations have led to ongoing international efforts to “deworm” school-aged children with either benzimidazole anthelmintics for soil-transmitted helminths or praziquantel for schistosomiasis, or both [15,19]. It is anticipated that the benefits of mass drug administration and deworming will also translate to sustainable poverty reduction [15,19,25]. However, rapid

Table 1
Poverty-promoting conditions with orphan vaccine programs in early research and development

Disease	Burden (DALYs) ^a	Chronicity	Child health	Maternal or perinatal health	Worker productivity	Selected references
Helminth infections						
Hookworm	22.1 million	++	++	++	++	[26,84–87]
Onchocerciasis	0.5 million	++	–	–	++	[89–91]
Schistosomiasis	4.5 million	++	++	++	++	[27,92]
Protozoan infections						
Amebiasis	NA ^b	+	+	+	+	[101–104]
Chagas	0.7 million	++	+	+	++	[106–112]
Leishmaniasis	2.1 million	++	++	++	++	[113]
Malaria	46.5 million	++	++	++	++	[11]
Bacteria infections						
Buruli ulcer	NA ^b	++	+	+	++	[118]
Chlamydia infections	5.9 million	++	+	+	++	[64,120–122]
Leprosy	0.2 million	++	+	+	++	[117,123]
Leptospirosis	NA ^b	+	+	+	+	[124]
Treponematoses	4.2 million	++	++	+	+	[125]
Tuberculosis	34.7 million	++	++	++	++	[12]
Viral infections						
Dengue	0.6 million	–	++	+	+	[126,127]
Japanese encephalitis	0.7 million	–	+	+	+	[127]
HIV/AIDS	84.5 million	++	++	++	++	[10]
Non-communicable disorders						
Drug use disorders	7.4 million	++	+	+	++	[31]

^a DALYs estimates obtained from either [9] (estimates for hookworm and schistosomiasis only) or [7].

^b Data not available.

post-treatment re-infection and the concerns about possible anthelmintic drug resistance have led to development efforts to complement school-based deworming with anthelmintic vaccines, such as for hookworm [26] and schistosomiasis [27]. While there is also substantial evidence for long-term neurocognitive impairments associated with *Plasmodium falciparum* malaria infection during childhood (reviewed in ref. [28]), other potentially vaccine-preventable neglected tropical diseases have not been studied extensively from this aspect. Of interest, however, is the finding that children with prenatal cocaine exposure exhibit long-term deficits in language development, academic performance, and intelligence [29,30]. Therefore, efforts to develop vaccines against cocaine and other drug addictions [31] could have important effects on this aspect of childhood development.

4. Impact on maternal and perinatal conditions

A strong body of evidence is emerging for the enormous impact of neglected tropical diseases on maternal–fetal outcomes including low birth weight, increased neonatal mortality, and maternal mortality. The most important tropical infections in terms of their impact during pregnancy are malaria (especially *P. falciparum* malaria), hookworm, schistosomiasis, Chagas disease, and syphilis. Approximately 50 million pregnant women live in malaria-endemic regions, where it is estimated that malaria is associated with 19% of infant low birth weight and 6% of infant deaths [32,33]. The association between hookworm and adverse consequences of pregnancy has been known since the early part of the 20th century [34]. Today, approximately 44 million pregnant women are infected with hookworm [35]. For both malaria and hookworm, much of the maternal and perinatal morbidity and mortality occurs as a result of the anemia caused by each agent [36,37]. Malaria is an especially important cause of anemia and its consequences in primigravidae, whereas hookworm is a greater problem among multigravidae [36,37]. Accordingly, treatment of malaria (e.g., intermittent preventive treatment and insecticide-treated nets) and hookworm (e.g., anthelmintic therapy) during pregnancy is associated with marked improvements in maternal and infant survival [32,38].

Other neglected tropical diseases also have important effects during pregnancy. Schistosomiasis caused by *S. mansoni*, like hookworm, was shown to increase the risk of anemia [39], and helminth co-infections caused by hookworms, schistosomes, and filariae were recently shown to be associated with increased risk of mother-to-child HIV transmission [40]. Chagas disease causes poor perinatal outcomes, including spontaneous abortion, fetal hydrops, and stillbirth [41], and there is a risk of maternal–fetal transmission that results in high rates of infant mortality and low birth weight [41–43]. Vertical transmission is increased in mothers with high parasitemia [44], and yet the available anti-trypanosomal drugs are contraindicated in pregnancy [42]. Maternal and congenital

syphilis remains a significant public health threat, with some estimates indicating that this condition results in greater than 500,000 fetal deaths yearly [45]. This number of deaths is equivalent to the deaths resulting from congenital HIV infection [45]. Globally, approximately 12 million adults are infected with syphilis [46]. In the United States, the case fatality rate for this condition is estimated to be approximately 6% [47]. Congenital leishmaniasis, on the other hand, is considered rare [48,49]. Cocaine use during pregnancy is linked to a number of adverse sequelae including intrauterine growth retardation, abruption placenta, pre-term labor [50,51], and low birth weight [52]. Maternal cocaine use is also linked with higher rates of adult syphilis, congenital syphilis, and HIV [53].

5. Impact on worker productivity

A third poverty-promoting feature of the neglected tropical diseases is their disabling character and therefore their impact on worker productivity. Among the vector-borne diseases, the limb deformities caused by dracunculiasis [54] and lymphatic filariasis [55–57], Chagas heart disease [58,59], leishmaniasis [60,61], and blindness caused by onchocerciasis [62,63] stand out for their economic impact, as does blinding trachoma [64].

One of the earliest features described for human hookworm infection was its impact on agricultural worker productivity. In many rural areas of the tropics, the prevalence and intensity of hookworm is highest (resulting in hookworm disease and anemia [18]) among agricultural workers. Thus, the major etiologic agent of human hookworm infection, *Necator americanus*, was first described by Bailey K. Ashford who went to Puerto Rico during the Spanish-American War and linked the parasite to anemia in the *jibaros* working on the sugar plantations [65,66]. Subsequently, hookworm was linked to reductions in working capacity and efficiency among laborers throughout the tropical regions of the Americas [67,68], Africa, and Asia [69]. More recent studies have confirmed the importance of soil-transmitted helminth infections on labor productivity, e.g., in banana plantation workers in St. Lucia [70], and in female tea pluckers in Bangladesh [71], although most of the evidence linking helminth infections to worker productivity is considered indirect [72]. Schistosomiasis and lymphatic filariasis also have a significant effect on the productivity of workers [73–75].

6. Impact on co-infections

In many cases it is difficult to ascribe injury and disability to a particular neglected disease because of their extensive geographic overlap. For that reason, it is common for an individual to be simultaneously affected by several of these conditions [8,13,76,77]. Such individuals are said to be *polyparasitized*. The geographic overlap of neglected

tropical diseases and the degree of polyparasitism is most commonly encountered in Sub-Saharan Africa [13], although this phenomenon also occurs frequently in Latin America [13,78] and in Asia. Therefore, the poverty-promoting potential of the neglected tropical diseases must consider the reality that they do not occur in isolation. This observation and the availability of drugs such as albendazole, azithromycin, ivermectin, and mebendazole, through large-scale donations from Pharma (or low-cost availability of praziquantel) provides the basis of designing “rapid-impact” packages for preventative chemotherapy [8,13,79]. Of concern, are the sustainability of this approach as a result of post-treatment re-infection and the possibility of emerging drug resistance.

The neglected tropical diseases also overlap with malaria [80] and HIV/AIDS [81]. Therefore, these two major killers are also occurring frequently in polyparasitized individuals [8]. The consequences of malaria, HIV/AIDS, and neglected tropical diseases co-infections were recently reviewed [8]. Anemia is predominant feature of the morbidity (and in some cases mortality) from malaria, hookworm and schistosomiasis, and there is a need to examine the additive effects of hemoglobin reduction from these conditions when they occur in combination. Moreover, there is evidence that helminths increase susceptibility to or worsen the progression of morbidity from malaria and HIV/AIDS (reviewed in refs. [8,81,82]). Therefore, the “other diseases” mentioned in MDG 6 are linked to HIV/AIDS and malaria, and there is a rationale for controlling neglected tropical diseases as a low-cost, effective mechanism for reducing the morbidity and mortality of malaria and AIDS [8]. In 10 Sub-Saharan African countries—Ethiopia, Ghana, Kenya, Mali, Malawi, Nigeria, Rwanda, Senegal, Tanzania, and Uganda, efforts are underway to bridge national control programs for malaria with those for neglected tropical diseases [83].

7. Antipoverty vaccines in the development of pipeline

The adverse consequences of the neglected tropical diseases and other conditions on child development, pregnancy outcome, worker productivity, and malaria and HIV/AIDS co-infections suggest a potentially important role for developing new generation neglected tropical disease vaccines

to combat poverty. In general, antigen discovery and pre-clinical development of neglected disease vaccines have not progressed much beyond early development. There are two major reasons for this situation. First, for eukaryotic pathogens, such as protozoa and worms, it has not been generally possible to exploit high throughput reverse vaccinology approaches because of the requirements for complicated eukaryotic expression vectors and animal models for vaccine testing [84]. Second, clinical development has not progressed for many of the antipoverty vaccines because of the absence of commercial markets and, therefore, industry interest. The few success stories in the area of neglected tropical disease vaccine development have required the establishment of new institutions and financing mechanisms including public–private partnerships (PPPs) and product development public–private partnerships (PD-PPPs or PDPs) [85]. Here we briefly summarize the current developmental status of some of the major antipoverty vaccines, including the role of selected PPPs and PDPs in establishing a development pipeline.

7.1. Vaccines for helminth infections

Two anthelmintic vaccines are in human clinical trials, one for human hookworm infection (‘hookworm’) caused by *N. americanus*, and the other for schistosomiasis caused by *S. haematobium* (Table 2). Together, hookworm and schistosomiasis account annually for approximately 345,000 deaths and 26.6 million DALYs in developing countries [8].

7.1.1. Hookworm

Hookworm occurs in an estimated 576 million people worldwide, with the greatest number of cases in rural impoverished regions of Sub-Saharan Africa, Asia and tropical regions of the Americas. *N. americanus* is the major species of hookworm worldwide. Most of the morbidity from hookworm is attributed to anemia and protein malnutrition [18]. The high rates of hookworm post-treatment re-infection, the diminished efficacy of currently available benzimidazole anthelmintic drugs with increased and frequent use, and the prospect of emerging drug resistance (reviewed in ref. [26]), have prompted a search for effective hookworm antigens. An antigen secreted by all species of infective hookworm larvae, known as ASP-2 (Ancylostoma secreted protein 2) was

Table 2
Human recombinant and conjugate antipoverty vaccines in clinical trials

Disease/etiologic agent	Vaccine/organization	Testing sites
Cocaine addiction	TA-CD/Xenova Group Limited, Cambridge, UK	Not specified
Hookworm infection/ <i>Necator americanus</i>	<i>Na</i> -ASP-2 Hookworm Vaccine/Sabin Vaccine Institute, Washington, DC, USA	Latin America: Brazil
Leishmaniasis/ <i>Leishmania</i> spp.	Leish-111f-MPL [®] -SE/Infectious Disease Research Institute, Seattle, WA, USA	Latin America: Brazil, Peru
Nicotine addiction	NicVAX [®] /Nabi Biopharmaceuticals, Boca Raton, FL, US	United States
Schistosomiasis/ <i>Schistosoma haematobium</i>	TA-NIC/Xenova Group Limited, Cambridge, UK	Not specified
	Sh28GST/Institut Pasteur, Lille, France	Sub-Saharan Africa, Niger, Senegal

selected for further development and testing based on a number of criteria established during preclinical testing in dogs and hamsters, and immunoepidemiological studies in humans (reviewed in refs. [26,86]). Anti-ASP-2 antibodies inhibit hookworm larval invasion in vitro and it is hypothesized that the vaccine prevents larvae from either reaching their final target organ, the small intestine, or from developing into adult hookworms [87]. The *Na*-ASP-2 Hookworm Vaccine was developed by the Human Hookworm Vaccine Initiative, a Washington, DC, PDP based at the Sabin Vaccine Institute (<http://www.sabin.org/hookworm.html>). The vaccine is comprised of a 21.3 kDa recombinant antigen cloned from *N. americanus* larvae, expressed by the yeast, *Pichia pastoris* and adsorbed to Alhydrogel[®] [26,87]. The *Na*-ASP-2 Hookworm Vaccine has undergone Phase 1 clinical testing in healthy human volunteers at doses of 10, 50, and 100 µg, and plans are underway to conduct subsequent clinical tests in hookworm-infected adults and children leading to proof-of-concept for its efficacy in Brazilian school-aged children [26,85–87]. Additional planning is in progress to formulate *Na*-ASP-2 with a second adult-stage antigen [26,86], as well as efforts to sustain the development and testing of the vaccine through partnerships with Instituto Butantan, a state-owned Brazilian vaccine manufacturer, the Oswaldo Cruz Foundation, and use of the vaccine by linking anthelmintic vaccination with deworming [26,85,86].

7.1.2. Onchocerciasis

Onchocerciasis is a vector-borne filarial infection and a major cause of blindness in the developing world. Approximately 18 million people are infected, with 99% of the cases in Sub-Saharan Africa. The current strategy for control relies on mass drug administration (MDA) of ivermectin, which targets the microfilarial stages of the parasite. However, there are long-term concerns about the sustainability of ivermectin MDA and the possibility of emerging drug resistance [88]. Efforts are in progress to identify potential vaccine antigens from *Onchocerca volvulus* [89,90], including an *O. volvulus* protein with homology to ASP-2 [91], but no development projects are underway.

7.1.3. Schistosomiasis and other platyhelminth infections

Schistosomiasis is a blood-fluke infection occurring in approximately 200 million people worldwide, with the majority of infections in Sub-Saharan Africa and Brazil. The two most important schistosomes causing human disease are *S. haematobium* (urinary schistosomiasis) and *S. mansoni* (intestinal and liver schistosomiasis). The disease burden of schistosomiasis has recently undergone re-evaluation with new data highlighting the chronic sequelae that results from deposition of parasite eggs in the bladder, intestine, and liver [17]. *S. haematobium* infection is particularly burdensome because of the large number of cases of hydronephrosis, renal failure, and bladder cancer that occurs in Sub-Saharan Africa. Despite the availability of praziquantel to treat this

condition, concerns about re-infection and the possibility of drug resistance has prompted efforts to develop a vaccine, with emphasis on developing a vaccine that reduces female worm fecundity, egg deposition, and egg viability [27]. Based on preclinical testing in mice and primates, and a demonstration in humans that antigen-specific IgG₃ correlates with age-dependent decreases in egg output, a 28 kDa *S. haematobium* glutathione-S-transferase (Sh28GST) was selected for further development by a group based at the Institut Pasteur in Lille, France [27]. In human trials, two alum-formulated injections of 100 µg of recombinant Sh28GST (produced in yeast) was shown to be safe and immunogenic with high levels of IgG₁ and IgG₃ [27]. Studies leading to proof-of-concept for Sh28GST in Senegal and Niger are in progress, with long-term plans to integrate the vaccine with existing chemotherapy-based control programs [92]. Additional vaccine antigens are also entering the development pipeline. First generation veterinary vaccines have been developed against liver fluke infection caused by *Fasciola hepatica* [93] and the cestode infections—cysticercosis and echinococcosis [94], although the corresponding human vaccines have not been developed.

7.2. Vaccines for neglected protozoan infections

Proof-of-concept has been established in animals for vaccine protection against *Entamoeba histolytica* (amebiasis), *Trypanosoma cruzi* (Chagas disease) and *Leishmania* spp. (leishmaniasis), the etiologic agents of the major poverty-promoting neglected protozoan infections. Together, Chagas disease and leishmaniasis account for approximately 100,000 annual deaths and 2.8 million DALYs [7,95], while the global disease burden from amebiasis has not been determined. Genome projects have been completed for *E. histolytica* [96], *T. cruzi* [97], and *Leishmania major* [98]. However, vaccine development for these diseases has been hampered by the absence of high throughput reverse vaccinology approaches, which led to the successful development of some anti-bacterial vaccines [84]. Among the reasons for this situation is the absence of a simplified eukaryotic expression system, which can simultaneously express hundreds of antigens, and the absence of suitable animal models for testing these antigens [84]. Recently, however, 100 novel vaccine candidates against murine *L. major* infection were studied in a high throughput screen [99]. Of the three diseases listed above, only the leishmaniasis vaccine has progressed to human clinical testing (Table 2).

7.2.1. Amebiasis

The highest disease burden from amebic colitis and liver abscess occurs in Mexico, Central and South America, Africa, and India. In Mexico it was estimated that approximately 1 million cases occur annually with 1216 deaths [100]. Several approaches have been taken for developing vaccines against amebiasis (reviewed in ref. [101]). Attenuated *E. histolytica* strains have been developed including a

strain that underwent silencing of an amebapore A virulence factor [102], and it is hoped that the recent completion of the *E. histolytica* genome [96] will make it possible to identify additional virulence genes and to conduct comparisons of *E. histolytica* with *E. dispar*, a non-pathogenic but morphologically identical amoeba [101]. Several promising subunit vaccines have undergone preclinical development. Based on studies in Bangladesh showing that individuals with secretory IgA antibodies to a Gal/GalNAc *E. histolytica* lectin exhibit a reduced risk of acquiring new infection [103], and studies showing that the native lectin is a protective antigen in gerbils [104], there has been interest in developing a subunit vaccine by testing recombinant proteins derived from the sequence of the 170 kDa lectin subunit [101]. Other candidate antigens have also been identified [101]. No PDPs have been created for developing a vaccine for amebiasis, nor combination anti-diarrheal vaccines that might provide protection against amebiasis together with other enteric infections [101].

7.2.2. Chagas disease

Chagas disease is a zoonosis caused by *T. cruzi* and transmitted by blood-feeding triatomine insects. Chronic infection can lead to a severe and debilitating cardiomyopathy, and a gastrointestinal condition those results in megaesophagus and megacolon. Approximately 10–12 million people are infected with *T. cruzi*, almost all of whom live in rural and impoverished regions of Central and South America, with approximately 45,000 deaths annually [95]. The two major drugs available for the treatment of Chagas disease are nifurtimox and benznidazole. However, the side effects of these drugs are severe and there is no strong evidence that even these treatments affect the progressive clinical development of cardiomyopathy or gastrointestinal pathology [105]. Both CD8+ cytotoxic T cells and IFN γ have been implicated in mediating resistance to Chagas disease [106,107], and the antigens encoded by the *T. cruzi* sialidase/trans-sialidase gene superfamily have been identified as targets of these responses [108]. Immunization protocols using purified *T. cruzi* proteins have been shown to protect mice against either infection or death [107]. Candidate proteins include a paraflagellar rod protein and a kinetoplastid membrane protein [107], and there has been some success in mice with DNA vaccines encoding trypomastigote surface antigen 1, LYTI, and a pool of trans-sialidase genes [107–110]. Even though the trans-sialidases are attractive vaccine targets because they are the immunodominant targets of CD8+ T cells, the large size of the trans-sialidase gene family and the potential for antigen variation may preclude their further vaccine development [111]. Both the genome [97] and proteome [112] of *T. cruzi* have been completed, opening the way for possible reverse vaccinology approaches [84]. No programs to develop human vaccines against Chagas disease are in progress.

7.2.3. Leishmaniasis

Leishmaniasis is a sandfly-transmitted kinetoplastid parasite of multiple different species causing cutaneous (CL),

mucocutaneous (ML) and visceral (VL) forms of the infection. The overall prevalence is approximately 12 million cases per year with an incidence of approximately 0.5 million cases of VL. VL is a major cause of morbidity and mortality in East Africa, India (especially Bihar State), North Africa, Southern Europe, and Brazil; it is also an opportunistic infection in patients with HVI/AIDS. The standard chemotherapy for VL is an antimony compound, which has significant toxicity and is associated with drug resistance in Bihar State and other regions. After inoculation in humans by the bite of a sandfly, the organism enters macrophages where it replicates as the amastigote form. Host immunity is mediated by T cells [113]. Leishmanization, immunization with live organisms obtained from lesions of patients suffering from CL, has been practiced for centuries and antedates vaccination—it is still practiced in parts of Central Asia [113]. The process has been refined using killed parasites or crude antigens in the presence of BCG, although evidence for the efficacy of such first generation vaccines is lacking. A second-generation recombinant vaccine has been developed through the Leishmaniasis Vaccine Initiative based at the Infectious Diseases Research Institute (IDRI) in Seattle, Washington (<http://www.idri.org/rd/leish>). The Leish 111f vaccine is a polyprotein containing three priority candidate antigens, TSA, LmSTII and LeIF fused in tandem. Formulated with 20 μ g of MPL[®] (4'-monophosphoryl lipid A from *Salmonella minnesota*), the Leish-111f-MPL[®]-SE vaccine affords protection in Balb/c mice for more than 14 weeks and has been extensively tested for safety in animals [113]. In humans, safety and immunogenicity has been demonstrated at antigen doses of 10, 20, and 40 μ g, and plans to evaluate the vaccine in therapeutic trials of infected patients with ML in Peru and CL in Brazil [113]. These studies are being evaluated in combination with standard chemotherapies in order to develop shorter and safer therapeutic regimens [113].

7.3. Vaccines for neglected bacterial infections

The major poverty-promoting bacterial infections include Buruli ulcer, *Chlamydia trachomatis* infections (including trachoma), leprosy, leptospirosis, and the treponematoses (including syphilis). Together, the Chlamydia infections, leprosy, and syphilis account annually for approximately 172,000 deaths and 10.3 million DALYs [7]. There are no disease burden estimates for Buruli ulcer or leptospirosis. The proteome for *C. trachomatis* [114], *Treponema pallidum* [115], *Leptospira interrogans* [116], and *Mycobacterium leprae* [117] has been analyzed for potential vaccine candidates, and some human trials have been conducted using earlier generation killed whole organism or live attenuated vaccines. Two PDPs have been established to develop and test and human neglected bacterial infections vaccines, including a joint initiative between IDRI and the American Leprosy Mission for a leprosy vaccine (http://www.leprosy.org/CC_Vaccine.html), and a second initiative between IDRI and GlaxoSmithKline

for the development of a vaccine against *C. trachomatis* (<http://www.idri.org/rd/chlam>).

7.3.1. Buruli ulcer

Buruli ulcer (named for the Buruli district in Uganda) is a disfiguring and debilitating cutaneous infection caused by *Mycobacterium ulcerans*. The full extent of Buruli's disease burden is unknown because it occurs primarily in remote and rural areas of West Africa [118]. The infection has also been described elsewhere in Africa, Asia, and the Americas. The organism is transmitted by water-borne contact through broken skin causing a disease that begins as a painless skin nodule and progresses to a necrotizing ulcer, usually on the extremities [118]. Children comprise approximately 50% of the diagnosed cases [118]. Early stage nodules can be excised, but once the disease progresses extensive surgery of the dead tissue and skin grafting is required. Antimicrobial therapy is often not successful. Vaccination with BCG produces significant protection against Buruli ulcer, but it is short-lived [118]. Efforts to improve this situation include booster immunizations with BCG, and the development of attenuated *M. ulcerans* vaccines [118]. Recent discoveries include the identification of a secreted family of three *M. ulcerans* secreted proteins (known as Ag85), and a unique mycobacterial toxin that may have a role in disease pathogenesis [118]. In principle, genes encoding synthetic enzymes for the toxin could be deleted in order to produce an attenuated strain of *M. ulcerans* [118]. The Global Buruli Ulcer Initiative, a partnership of Member states, non-governmental organizations, research institutions, and the WHO, has been established to develop better control tools for the treatment of Buruli ulcer (<http://www.who.int/buruli/en>), however, no major effort is underway to develop a vaccine for human testing.

7.3.2. Chlamydia infections

C. trachomatis causes several poverty-promoting conditions in humans. Trachoma (serovars A, B, Ba, and C) results when chronic ocular infection causes in turned eyelashes leading to corneal scarring. Approximately 84 million cases occur worldwide, resulting in 8 million cases of visual impairment [119] (<http://www.trachoma.org>). *C. trachomatis* is also an important cause of sexually transmitted disease (STD) in developing countries (serovars D-K and L1-3), associated with urethritis, cervicitis, and lymphogranuloma venereum, and accounting for approximately 20% of the estimated 500 million STDs worldwide [120]. Infection during pregnancy can be associated with vertical transmission and neonatal conjunctivitis and pneumonia. The cornerstone of endemic trachoma control has been the so-called SAFE strategy comprised of eye surgery, mass drug administration with a once-yearly dose of azithromycin, face-washing, and environmental changes to improve water and sanitation (<http://www.trachoma.org>). The ideal vaccine would be one that protects against multiple serovars and prevent chronic sequelae, including blindness, pelvic inflammatory disease, and infertility [121]. Chlamydia immunity relies on eliciting

strong Th1 responses [119,120]. During the 1960s, Thomas Grayston and his colleagues at the University of Washington (Seattle, WA) as well as other groups in the UK and Italy conducted several human trials with experimental whole cell vaccines (both killed and living vaccines) derived from chicken egg yolk sac [121]. These vaccines were expensive to produce and gave only limited and short-term protection; in some cases they worsened the course of the disease [121]. Because it is currently not feasible to genetically modify chlamydiae in order to produce safe attenuated strains, the recombinant subunit approach is the one considered most likely to be successful [120]. Among the candidates showing the greatest promise is a 40 kDa outer membrane antigen, known as MOMP, as well as the polymorphic outer membrane proteins (POMP), the PorB family of membrane antigens, an ADP/ATP translocase, a *pgp3* plasmid protein, and others [120,121]. Several of these antigens are undergoing preclinical vaccine development at Aventis Pasteur, Antex, and IDRI together with GSK [121]. These candidates may also be effective when used together as a multisubunit vaccine or as DNA vaccines [120]. No human trials are in progress [122].

7.3.3. Leprosy

Leprosy is a chronic and disabling granulomatous infection caused by *M. leprae*. Through widespread use of multidrug therapy (MDT), the disease burden caused by leprosy has been reduced more than 90%, with approximately 410,000 new cases reported in 2004 (<http://www.leprosy.com>). Leprosy has been eliminated in all but 9 out of the original 122 countries where leprosy was first noted to be a public health problem in the 1980s (<http://www.who.int/mediacentre/factsheets/fs101/en/>). Today, most of the existing and new cases occur in India. There is cautious optimism that leprosy could be eliminated through continued and aggressive use of MDT. However, there has also been some interest to develop anti-leprosy vaccines. BCG has been shown to partially protect against leprosy [123], and through a joint initiative between IDRI and the American Leprosy Mission 5 potential vaccine candidates have been identified through an analysis of the *M. leprae* proteome (http://www.leprosy.org/CC_Vaccine.html). No clinical trials are in progress.

7.3.4. Leptospirosis

Leptospirosis is a zoonotic infection caused by pathogenic spirochetes (genus *Leptospira*). Human infection occurs through contact with water or soil contaminated by the urine of infected animals. The infection is common in much of the tropics and subtropics—in these areas it is common to find infectious foci in urban slums and among individuals who work in mining and other high-risk populations. Approximately 10% of patients become severely ill and develop Weil's disease characterized by high fever, jaundice, and hemorrhagic sequelae. Veterinary vaccines have been developed for cattle and dogs. In the 1930s, a heat-killed whole cell vaccine from leptospiral cultures was developed in Japan

where it was shown to protect coal miners from Weil's disease [124]. A serovar-specific formalin-killed whole cell leptospira vaccine is still available in Japan, and elsewhere [124]. To date, each vaccine is serovar specific. A number of immunogenic proteins have been identified from the outer membrane of pathogenic leptospira, some of which can generate cross-protection against different serovars when used as subunit vaccines [124]. The completion of the genome for *L. interrogans* may accelerate development through reverse vaccinology [116].

7.3.5. Syphilis and other treponematoses

The human treponemal infections include, yaws (a disfiguring childhood infection caused by *T. pallidum pertenue*, endemic to Central America, the Caribbean, equatorial Africa, and equatorial islands of Southeast Asia), pinta (a disease of young adults in the Americas caused by *T. carateum*), bejel or endemic syphilis (a disease of children living in nomadic families on the Arabian peninsula and on the southern border of the African Sahara desert, caused by *T. pallidum endemicum*), and venereal syphilis (*T. pallidum pallidum*). The global burden is approximately 25 million cases, the majority of which occur in Sub-Saharan Africa and Asia [125]. Although little is known about the mechanisms of immunity to syphilis there have been some efforts to develop syphilis vaccines. Both whole cell killed vaccines and live attenuated vaccines have been developed, although scale-up of these vaccines has not been feasible because it is not yet possible to culture these organisms in vitro [125]. Several investigators have identified putative outer membrane proteins as potential surface-exposed vaccine antigens, and to test them in rabbits and other animal models. No human trials with recombinant antigens are in progress.

7.4. Antiviral vaccines

Many investigators consider the major arthropod-borne flavivirus infections, namely yellow fever, dengue types 1–4, Japanese encephalitis, West Nile virus, and tick-borne encephalitis as important neglected tropical diseases. However, their acute presentation and the observation that these are largely emerging infections places this group in a distinct category from the other neglected tropical diseases, which are poverty-promoting by virtue of their chronicity and their impact on child development, pregnancy, and worker productivity. New developments in flavivirus vaccines were recently reviewed [126], as well as a study of the cost-effectiveness of a pediatric dengue vaccine [127]. In 2003, the Pediatric Dengue Vaccine Initiative was established at the International Vaccine Initiative in Seoul, Korea (<http://www.pdvi.org>).

7.5. Vaccines against drugs of abuse

Drug addiction to cocaine and tobacco are major problems worldwide. For cocaine abuse, there is no proven pharmacological therapy. As a result, several lines of approaches

have been attempted to produce anti-cocaine conjugate vaccines comprised of a cocaine hapten, a linker, and a protein. An anti-cocaine vaccine known as TA-CD (produced by Xenova Group Limited, Cambridge, UK) is in Phase 2 clinical trials [31]. The vaccine is comprised of norcocaine acylated with succinic anhydride and conjugated to bovine serum albumin [31]. A Phase 1 study demonstrated that three intramuscular injections of TA-CD were well tolerated, with anti-cocaine antibody levels peaking after three months [31]; Phase 2 studies showed that maximum mean antibody responses occur between 70 and 90 days post-vaccination with cocaine-specific antibodies persisting for at least 6 months (<http://www.xenova.co.uk/dc.ta.cd.html>). Similarly, several groups are developing nicotine vaccination strategies. The Tobacco Free Initiative (TFI) of the World Health Organization estimates that tobacco is responsible for the death of 1 in 10 adults worldwide, resulting in 5 million deaths annually (<http://www.who.int/tobacco>). Two vaccines against nicotine addiction are in clinical trials [31], including a Phase 2 clinical trial for NicVAX[®] (Nabi Biopharmaceuticals in Rockville, MD, USA, <http://www.nabi.com>) comprised of nicotine linked to carrier protein recombinant *Pseudomonas aeruginosa* exoprotein A, and the other, a nicotine–cholera toxin B immunoconjugate (TA-NIC) under development by Xenova, which has completed two Phase 1 studies (<http://www.xenova.co.uk/dc.ta.nic.html>).

8. Developing the antipoverty vaccines: moving candidates through the pipeline

Of the 11 neglected, poverty-promoting infections described, e.g., hookworm, onchocerciasis, schistosomiasis, amebiasis, Chagas disease, leishmaniasis, Buruli ulcer, Chlamydia infections (trachoma), leprosy, leptospirosis, and syphilis and the other treponematoses, vaccine projects for only three diseases (e.g., hookworm, schistosomiasis, and leishmaniasis) have progressed to the point where a recombinant vaccine has been developed, approved by an international regulatory body, and clinical testing in either Phase 1 or 2 trials have commenced. Ironically, we have less baseline information in terms of a genomic and proteomic database for hookworm and schistosomiasis than we do for most of the other infections for which no human clinical trials are underway. For *E. histolytica*, *T. cruzi*, *L. major*, *C. trachomatis*, *L. interrogans*, *M. leprae*, and *T. pallidum*, the genome has been completed; for some of these agents proteomic analysis for potential vaccine antigens has commenced. Therefore, although there are technical hurdles that confront vaccine development projects for all of the 11 poverty-promoting tropical infections, these no longer appear to be the rate-limiting factor. Instead, our technical ability to produce neglected disease vaccines has outpaced the social and political will needed to translate scientific discoveries into products.

Most of the vaccines in Table 1 would be considered “orphan vaccines” by the major regulatory agencies for health

products in the U.S., Europe, and Japan. U.S. legislation provides orphan status to diseases of fewer than 200,000 Americans, or diseases that have no potential recovery costs from U.S. sales [128]. European and Japanese orphan status requires a prevalence of less than 5 per 10,000 Europeans and 2.5 per 10,000 Japanese, respectively [128]. Orphan status also requires that no current methods of prevention or treatment exist. For at least three potential neglected disease vaccine projects, those for the STDs caused by *C. trachomatis*, *T. pallidum*, and leishmaniasis there is a small but significant commercial market in North America, Europe, or Japan. This is also true for the addiction vaccines.

In order to examine the challenge of developing orphan vaccines for small commercial markets, several groups have looked at the prospects of establishing markets for vaccines where none existed previously. Efforts have focused on creating advanced market commitments, which include price guarantees and a market size of approximately \$3 billion per disease (reviewed in ref. [129]). This approach relies heavily on a specific paradigm, namely a mechanism to provide incentives for the larger vaccine manufacturers, with particular emphasis on vaccines to combat HIV/AIDS, malaria, and tuberculosis (the “big three diseases”). The role of such advanced market commitments for the neglected tropical disease vaccines outlined above is less clear. These diseases do not have the same high profile as the big three diseases and it may not be feasible to implement advanced commitments for them. Instead, it was the establishment of the PDP that led to the successful development and testing of vaccines for hookworm, leishmaniasis and schistosomiasis. In each case, the PDP was headed by a champion with a track record of research on that particular disease of interest (and in so doing developed an antigen pipeline). This led to efforts to identify funds and shift the focus of the investigator’s laboratory from basic research to vaccine development [85].

9. Global access for the antipoverty vaccines

Early development of neglected disease vaccines requires a program for its long-term and sustainable development. In the case of the *Na-ASP-2* Hookworm Vaccine, an important strategy is to pursue vaccine manufacture through partnerships with innovative developing countries (IDC). IDCs are middle-income countries, such as Brazil, Cuba, China, and India, with modest economic productivity but which have nonetheless achieved a high level of innovation in biotechnology (evidenced by their per capita numbers of peer-reviewed papers, international patents, and production of drugs, vaccines, and diagnostics) [130]. Many of the IDCs are highly endemic for the major neglected tropical diseases; this affords an opportunity to partner with IDCs not only for purposes of vaccine development but for clinical testing for efficacy, as well. Therefore PDP-IDC liaisons represent a promising avenue for the development and testing of the antipoverty vaccines [85].

Another unique feature of the poverty-promoting diseases is that many of them affect older children, adolescents, and young adults. Therefore, many of the antipoverty vaccines may be developed and used for individuals outside of the traditional infantile period and therefore outside of the Expanded Program on Immunization. This situation will require innovation in order to develop field vaccination programs and introduce vaccines into alternative age populations.

Some of these orphan products could be developed as school-based vaccines, especially in association with mass drug administration programs, such as those currently in place for deworming for soil-transmitted helminth infections and schistosomiasis. This includes the possibility of developing vaccine-linked chemotherapy initiatives in which the two interventions are used in a complementary manner [92]. School-based vaccination has also been proposed for recently developed cervical cancer vaccines, and this concept could also be expanded for some of the other STD antipoverty vaccines (e.g., Chlamydia infection and syphilis). Antenatal vaccination is another promising venue.

It is expected that the successful development, testing and distribution of the antipoverty vaccines will require carefully done cost-effectiveness analyses in order to justify the establishment of new paradigms and strategies for health products and their integration into developing country health systems. There is also a potential concern for product saturation, meaning the simultaneous development and distribution of more products than could be handled by current health infrastructures. An additional hurdle will be to identify mechanisms by which the Global Alliance for Vaccines and Immunization (GAVI) could accommodate these new vaccines. The Harvard economist David Canning argues that interventions against neglected tropical diseases represent important investments in human capital [131], a concept that promises to become an important global health theme in the coming decade.

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