Lepr Rev (2007) **78,** 338–342

Is there a role for a vaccine in leprosy control?

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Accepted for publication 14 November 2007

'Morbidity control' is a recurring theme of a recent WHO report entitled "Global strategy for further reducing the leprosy burden and sustaining leprosy control activities: 2006–2010".[2] While the approach seems rational, based on our current tools for controlling leprosy (i.e., timely detection of new cases, treatment with effective chemotherapy, prevention of disabilities and rehabilitation), it also seems resigned to accepting a future with leprosy. Should we accept the 'morbidity control' paradigm or are there other approaches to be pursued that may provide new insight into leprosy control and possibly lead to interventions moving us along the path to eradication?

I think most agree it is prudent to continue to support those strategies that are in place today but to insist on stringent evaluation of leprosy services and their results to ensure that integration of leprosy services is having the desired effect on the disease. Concomitantly, we must continue to pursue the search for new tools that, when applied appropriately, inform control practices leading to improved services and, eventually, the eradication of leprosy. At the top of my list of areas for continued and renewed study are the subjects of nerve damage, transmission and vaccines. In this commentary I will focus on the subject of vaccines. I will describe recent advances that are driving renewed interest in vaccine development for pathogens like *M. leprae* as well as address when and where a leprosy vaccine might be applied in a global strategy to eradicate leprosy.

Technological advances

Large-scale genome sequencing projects of pathogenic bacteria have led to new insights regarding mechanisms of virulence and drug resistance, metabolic capabilities under varying environmental growth conditions and genetic variation responsible for pathogen emergence. With regard to vaccine development for leprosy the explosion of genomic information from both the pathogen (*M. leprae*) and its natural hosts (man and armadillo), has been revolutionary. In addition, the speed with which antigen discovery can proceed today has

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been increased by an order of magnitude. Enhanced antigen discovery has occurred due to a convergence of procedures for *in silico* identification of potential protein antigens and gene cloning and recombinant protein expression tools that can be used for subsequent immunologic characterisation and vaccine efficacy trials of high-priority antigens.

Brennan *et al.* were the first to report the overall protein make-up of armadillo-derived *M. leprae* by two-dimensional polyacrylamide gel electrophoresis.[12] Their results showed approximately 391 cell-associated proteins. This may be an underestimate of *M. leprae*'s proteome because purification procedures used to prepare the bacilli are not capable of capturing secreted proteins. *In silico* analysis[16] and gene transcription surveys (D. Williams, personal communication) of viable *M. leprae* suggest the presence of many previously unidentified *M. leprae* proteins. Among other characteristics, these proteins may encompass activities involved in virulence or encode peptide sequences capable of being immunogenic during various stages of infection. Among this group of transcribed genes are protein homologs to immunodominant *M. tuberculosis* antigens as well as proteins involved in DNA replication, cell division, SecA-dependent protein secretion, energy production, intermediary metabolism, iron transport and storage. This growing stockpile of potential antigens for study has refocused our attention on the tools needed for testing large numbers of new antigens with the intent of selecting those antigens that should go forward for vaccine efficacy testing.

One approach for winnowing the large number of protein antigens to manageable numbers is to demonstrate immunogenicity in patients. This can be done by demonstrating antibody, cell-mediated immune (CMI) responses, or both, *in vitro*. Recently, Slayden *et al.*[8] published a powerful approach for detecting serum antibody from leprosy patients utilising a protein array with approximately 30 *M. leprae* native antigens and recombinant proteins. This methodology could be scaled-up to screen all proteins from *M. leprae* using serum from patients representing one or more forms of leprosy. Proteins selected as immunogenic in this screen could then be prepared in bulk and tested in CMI assays for T-cell reactivity using blood cells from leprosy patients across the disease spectrum as well as their contacts. Proteins meeting defined criteria could then be tested in the mouse foot pad assay using only those adjuvants cleared for human use. Further experimental testing of leprosy vaccines in animals may not be necessary beyond the mouse. However, recognising the limitations associated with the mouse foot pad model (e.g., limited growth of *M. leprae* and uncharacteristic pathology) begs the question; should we be testing our best candidate vaccines in the armadillo, a full disease spectrum animal model of leprosy?

Renewed interest in the armadillo model of leprosy has been sparked by the recent acquisition of its genome sequence (http://www.broad.mit.edu/ftp/pub/assemblies/mammals/armadillo). Truman *et al.* have begun the task of identifying, cloning and expressing armadillo genes that can be used to develop assays for monitoring aspects of leprosy pathogenesis and corresponding immune responses.[1] Among the genes first reported have been IL2, TNFa and IFNg (R. Truman, The armadillo model: a full spectrum host, US-Japan Cooperative Medical Sciences Program, NIAID, 40 th TB and Leprosy Research Conference, Seattle, WA, 2005), all of which are important mediators of an effective CMI response. Further exploitation of the armadillo model at the cell and molecular level could uncover critical immunological parameters associated with protection from, and susceptibility to, infection around which a potent vaccine could be configured and tested. Testing for vaccine efficacy in the armadillo would also allow for assessment of efficacy across the disease spectrum as the armadillo manifests similar bacterial growth characteristics and histopathological features observed in humans infected with *M. leprae*.[14]

The potential for technological advances in vaccine formulation for leprosy can be seen in the latest iteration of experimental TB vaccines.[3] Delivery platforms for this new generation of TB vaccines take the form of subunit proteins with novel adjuvants, recombinant vectors, including modified vaccinia Ankara (MVA) or adenoviruses and genetically modified mycobacteria, such as *M. tuberculosis* auxotrophs or recombinant BCG over-expressing *M. tuberculosis* proteins. Some have been shown to be more potent than standard BCG vaccines in animal models of tuberculosis and would likely be administered in concert with BCG in humans. Viral and recombinant BCG vaccines overexpressing *M. tuberculosis* protein(s) take advantage of the vector's ability to induce strong CD4 and CD8 responses, a bonus effect often seen with live vaccines. A drawback sometimes associated with live vaccines, however, is the potential for inducing disseminated infections following vaccination of individuals with immunological impairment, in particular, severe-combined immunodeficiency disorder (SCID), chronic granulomatous disease and AIDS. This underscores the need for thorough infection-toxicity testing of new live vaccines in animal models lacking both arms of the immune system. Similarly, implementation of new, live vaccines in humans must be accompanied by systematic evaluation of vaccine recipients for immune compromising conditions prior to vaccination. It is important to remember that disseminated BCG infections due to vaccination in healthy immunocompetent neonates is rare but the risk of disseminated BCG disease is increased several hundred-fold in HIV-infected infants compared to the documented risk in HIV-uninfected infants.[9]

Subunit protein vaccines avoid this potential dilemma. However, significant immune stimulation generally requires multiple injections. Another approach is to deliver a sub-unit protein vaccine subsequent to an earlier immune stimulus. This approach is often referred to as 'prime-boost' and is based on our classical understanding of the kinetics of the immune response in humans. In the case of TB vaccines the 'prime-boost' approach will likely be a combination of neonatal BCG vaccination followed by a second vaccine, either a subunit protein or other biologic. It remains debatable as to when the booster should be given to provide an efficacious outcome. Arriving at such an answer is often determined empirically through vaccine studies in humans where multiple combinations and permutations can be tested. But in the case of a low incidence disease like leprosy, multiple arms of a vaccine study are difficult to organise to address these issues. So, it may be worth considering the timing of a prime-boost strategy that would synergise immunological potency with epidemiological characteristics of disease risk that would result in the best chance to strengthen resistance to leprosy within the community, thereby limiting the spread of disease.

In the near term we should be exploring all of these vaccine platforms using both newly identified, as well as previously characterised, proteins in a quest for a potent and inexpensive vaccine for application in leprosy control. A practical approach for selecting antigens for a new leprosy vaccine would focus on 1) proteins shown to be immunogenic in a majority of leprosy patients and exposed contacts, 2) protein antigens known to be homologous with TB proteins, and 3) proteins shown to induce protective immunity in the mouse foot pad assay. The reasoning for selecting proteins with homologs in *M. tuberculosis* extends from the fact that *M. leprae*, *M. tuberculosis* and *M. bovis* BCG have, as part of their make-up, many proteins in common. If cross-reactivity between BCG antigens and *M. leprae* antigens can be exploited, this characteristic may provide the potential for enhanced vaccine efficacy when delivered in a prime-boost strategy with BCG. BCG vaccination is likely to remain in place in most TB-endemic countries because of its beneficial effects on tuberculomeningitis in children and its demonstrated impact in some areas against leprosy.[4]

Application of a leprosy vaccine

Given that innovation often precedes implementation of new products, it is important that we consider potential uses of new vaccines for leprosy as science drives the advances required to provide these tools. However, investing time and resources into developing a vaccine without a reasonable vision of how to use it is unwarranted and could be considered wasteful. So, what are some of the issues surrounding vaccines in leprosy and what might a leprosy control strategy look like with a vaccine component?

From a purely financial perspective it has been argued that vaccines are not cost-effective measures when applied universally to stop transmission of low incidence diseases. Leprosy certainly falls in the low incidence category; however, this argument doesn't preclude developing a post-exposure, therapeutic vaccine as part of a leprosy control strategy. A therapeutic vaccine could be offered to individuals in close contact with an index case and, therefore, thought to be at risk for infection. Because of BCG's reported efficacy as a vaccine against leprosy,[6,10,11,13,15] the Ministry of Health of Brazil recommends vaccination of household contacts of leprosy cases. This approach in essence constitutes a prime-boost or revaccination strategy with BCG in an attempt to bolster immunity to leprosy initiated with neonatal vaccination with BCG. Cunha *et al.* have begun a study to evaluate the impact of BCG revaccination on schoolchildren, some of whom may be contacts of leprosy cases.[5] The study is being conducted in 286 schools in Manaus, Brazil and includes over 150,000 school children, of whom over 70,000 are in intervention schools. BCG was given intradermally and follow-up is currently ongoing. It is anticipated that results from this study will lend support for or against revaccination as a means of further reducing leprosy in both household contacts as well as non-household contacts of leprosy patients in Brazil.

A therapeutic vaccine could also be used to supplement MDT of a newly diagnosed patient. Examples of the successful use of a therapeutic vaccine to enhance leprosy chemotherapy have been reported using *Mycobacterium w*.[7,17] While concern was raised with this approach, regarding increased appearance of Type 1 reactions in vaccinated individuals, neither increased neuritis nor type 2 reactions were observed in vaccinated patients.

If one accepts the potential use of a therapeutic vaccine under specific conditions (e.g., household contacts), can a wider role for vaccines in leprosy be envisioned? For example, could a vaccine strategy be organised that would enhance protection in those individuals at greatest risk for contracting leprosy who are not in close contact with a case of leprosy? This can be a significant percentage of new cases and has been estimated to approach 50% in some instances. While it could be argued that this is when a universally applied vaccine would be appropriate, the cost of vaccinating all citizens to prevent relatively few cases of leprosy is difficult for public health officials to rationalise. Accordingly, what is needed prior to implementing a broader-coverage vaccine is an improved understanding of host susceptibility factors for leprosy and a fuller picture of transmission patterns for this disease. A combined understanding of these two issues are critical to determining risk factors for contracting leprosy and provide public health officials and vaccinologists the information needed to implement new vaccine strategies, should they be called for in the quest for improved leprosy control and eventual eradication.

From a disease control perspective, vaccines are not unlike drugs in that they may be applied in a prophylactic (pre-exposure) or therapeutic (post-exposure) control strategy. Potent vaccines have the added advantage of producing long-lived immunological memory which can block multiple exposures over the host's lifetime. Thus, an effective prophylactic

vaccine for leprosy would break transmission by conferring immediate as well as extended protection to vaccine recipients from infection with *M. leprae*. A prophylactic vaccine should also protect against both drug-susceptible and drug-resistant strains, helping curb the emergence of drug resistance. Accordingly, reliance on secondary preventative measures, such as chemotherapy, is unlikely to advance us to the next level of disease eradication. In my view vaccines hold that promise.

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