Vaccines against bacterial zoonoses

Bacterial zoonoses such as anthrax, typhus and plague, have historically caused devastating human disease and have the potential to do so again should the appropriate environmental and social conditions arise. Others, including campylobacteriosis and salmonellosis are very common worldwide; some, such as brucellosis and leptospirosis, are of major economic significance because of their wide distribution and impact on animal production and public health. Emerging problems include Lyme disease associated with *Borrelia burgdorferi* and related species in Europe and North America, tick-borne relapsing fever associated with *Bor. hermsi* in the USA, ehrlichiosis and the expanding range of disease syndromes caused by *Bartonella* spp in immunocompetent and immunodeficient individuals.

Because of the need for contact with the animal host, its products or an arthropod vector, most human zoonoses are occupational, recreational or travel-associated. However, some are important for a more sinister reason: because of their high infectivity and ability to establish incapacitating or lethal infections from small inocula, some zoonotic agents are potentially useful as biological warfare agents. Indeed some, such as *Bacillus anthracis*, *Burkholderia mallei*, *Francisella tularensis* and *Yersinia pestis*, have been used for this purpose. Recognition of the inadequacies of currently available vaccines in the context of biological warfare has highlighted the need for new developments.

Vaccines are currently available for the prevention of anthrax, brucellosis, leptospirosis, plague, tularaemia, typhus and Q fever. Some of these have a very limited availability outside certain geographical areas and few are licensed in developed countries. The safety and efficacy of some of these products is also in question. Research on vaccines for zoonotic disease has been fairly limited, but the development of molecular cloning techniques has made possible the large-scale production of purified antigens and attenuated strains with defined genetic modifications.

Two types of anthrax vaccine for human use are available in the UK and USA. Both are based on partially purified protective antigen (PA) of the *B. anthracis* toxin complex adsorbed on to an aluminium adjuvant [1]. A live vaccine strain derived from the Sterne attenuated strain has been used in Russia and other countries of the former USSR [2]. The PA subunit vaccines are used to protect occupationally exposed groups and are evidently safe although transient adverse reactions seem frequent. Animal studies and epidemiological data provide circumstantial evidence of protection against respiratory and cutaneous anthrax in employees in high risk industries [3]. However, direct evidence of protective efficacy in man against aerosol challenge is lacking and experiments in animals suggest that protection against different *B. anthracis* strains may not be uniform [4]. The efficacy of this type of vaccine may be inherently less than that of live vaccines [2], but there would be considerable resistance to using the Russian vaccine in Western countries because of concerns over residual virulence and reactogenicity. Alternative options are limited. The key factors in the pathogenesis of anthrax are the production by the organism of the anti-phagocytic poly-D-glutamic acid capsule and the toxin complex. The latter comprises an oedema factor with adenyl cyclase activity [5], and a lethal factor that has metalloprotease activity and probably targets a macrophage subcellular protein [6]. Both toxins use PA for cell attachment and uptake, so that antibody to PA will neutralise both. Developments of highly purified PA for formulation with new adjuvants [7], or delivery of the antigen *in vivo* either in a ‘designer’ attenuated strain with defined genetic modifications or in a heterologous vector, have been effective. PA emulsified with monophosphoryl lipid A was more protective than PA with aluminium adjuvant [7] and cloning of the PA gene into an Aro-deletion mutant of *Salmonella enterica* serotype Typhimurium produced an expression system that stimulated immunity in mice to challenge by various routes, including respiratory aerosol exposure [8]. It is not yet known if these new vaccines will answer the need for a readily administered, low reactogenicity vaccine that will protect against all forms of anthrax.

Several vaccines protect against human brucellosis. The attenuated strains *Brucella abortus* S19 and *Br. melitensis* Rev 1 retain virulence for man and are unsuitable, but a variant of S19, *Br. abortus* S19-BA, has been used in the former USSR to protect occupationally exposed groups. Efficacy is limited and annual re-vaccination is needed. Adverse reactions are reported to be mild provided that the vaccine is administered by intradermal injection or skin scarification and not subcutaneously and that the recipients have not been sensitised. Another attenuated strain, *Br. abortus* 104M, is administered in China for similar purposes by the skin scarification and aerosol routes.
Neither of these vaccines would meet Western requirements for safety and efficacy.

A non-living vaccine with a phenol-insoluble peptidoglycan fraction [9] is claimed to stimulate a basic immunity to be reinforced by subclinical infection. Although of low reactogenicity, infections occurred in some recipients and its efficacy remains questionable. A protein-polysaccharide complex derived from smooth phase *Brucella* strains by mild acid hydrolysis is reported to be as protective as the *Br. abortus* 19BA vaccine, but less reactogenic, at least in individuals not sensitised by previous infection [10].

Current strategies for the development of improved vaccines against human brucellosis centre around the development of attenuated strains with defined genetic modifications. The RB 51 strain, which was developed for the immunisation of cattle [11], is unlikely to be useful in man as it is no more protective than S19 and its human pathogenicity is uncertain. Equivalent O chain deficient mutants of *Br. melitensis* or *Br. suis* may be worth examining but the most promising trend is the development of defined metabolic derivatives of virulent strains such as purE mutants [12, 13]. Because of the need to stimulate effective cell-mediated responses, subunit vaccines are less promising unless incorporating a delivery system or adjuvant capable of triggering Th1 responses. Fusion proteins incorporating the L7/L12 ribosomal proteins trigger antibody and cell-mediated responses and protection against challenge in mice [14]. They may have a future as components of human vaccines, possibly with detoxified lipopolysaccharide conjugates [15].

Current whole-cell plague vaccines stimulate immunity against bubonic plague, but are probably ineffective against the pneumonic disease [16, 17]. Subunit vaccines based on the fraction I and V antigens, either as purified proteins or fusion proteins, are effective against respiratory and parenteral challenge in animal models [18, 19]. Expression of these antigens in heterologous live vectors such as *Aro* mutants of salmonella also stimulates good protection [20] although attenuation may not be complete [21]. These vaccines have yet to be evaluated in clinical trials.

Vaccines against *Bor. burgdorferi*, based on the outer surface protein Osp A [22], are in clinical trial in the USA. As most infections in North America are caused by *Bor. burgdorferi sensu stricto* antigenic variation is not a problem, but in Europe infections are also caused by *Bor. afelli*, *Bor. garini* and unclassified species, in which the Osp A protein varies. Vaccines for Europe will have to be based on a cocktail of antigens; emphasis is being placed on the Osp B, Osp C, OspE and OspF proteins [23]. Such vaccines will probably include tick-borne fever antigens.

There seems much less interest in developing vaccines against *Bor. hermsi* and other relapsing fever spirochaetes even though these can cause lethal infections. This presumably reflects the perceived marketability of these vaccines, although relapsing fever vaccines would also be technically more difficult to develop because of antigenic variation in vivo.

A vaccine against tularaemia based on the LVS strain [24] is effective against the systemic as well as the localised oculo-glandular form of the disease, provided that the challenge is within the protection threshold. Immunity depends on a humoral response to the lipopolysaccharide and, possibly, a cell-mediated response to outer-membrane or other proteins [25]. Subunit vaccines may be feasible but the Fop A protein expressed in a salmonella vector was not protective [25].

A killed whole-cell vaccine based on phase 1 organisms is efficacious against occupational infection with Q fever [26]. An acid extracted subunit vaccine has also been developed [27], and a live attenuated strain (M44), producing predominantly phase 2 organisms, has been used in the former USSR [28].

Vaccines prepared from killed yolk sac-grown organisms are available on a local basis for typhus and the spotted fevers, but little work is being done on other rickettsial infections. Some of these are clearly emerging as significant problems and more emphasis on vaccine development would seem justified. *Orienta (Rickettsia) tsutsugamushi* is still a problem in some areas, but the wide range of serotypes makes vaccine development difficult.

Vaccines against leptospirosis are widely used in dogs, cattle, pigs and sheep and appear to be protective. Equivalent vaccines for protection against human disease are available in only a few localities, although recreational exposure through water sports is an increasing problem. The immunity stimulated by killed whole-cell vaccines is serotype-specific and some, preparations may have to contain seven or more serotypes to cover locally prevalent strains [29]. The variety of potential serotypes to be covered complicates formulation, but new approaches, such as nucleic acid vaccines, may overcome this problem.

Considerable progress is being made in the development of vaccines against some zoonoses. However, there is less activity against diseases that are perceived solely as Third World or minority problems. Economic factors are likely to continue to regulate progress in this area.

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References


