

SECTION 4

Regulatory and Ethical Aspects



Untitled – Ivo Saglietti
Photography

'I want no more than a hand,
A wounded hand, if possible.
I want no more than a hand,
even if I spend a thousand nights with no bed.'

Casida of the Impossible Hand (Poem)
– Federico Garcia Lorca

Ivo Saglietti

Genoa, Italy

Ivo Saglietti was born in 1948 in Toulon, France. He traded his movie camera for still photo equipment in 1978 and went to work in Paris for American and French agencies covering Latin America and the Middle East. In 1988, he began devoting his time to personal projects: taking photographs of Latin America, Kosovo, and the Mediterranean region's borders. He received the World Press Photo Award in 1992 and 1999 and the Enzo Baldoni prize in 2007 for 'Water and Oil', a photo essay on the Niger Delta. He lives in Genoa.

TB is a disease of poverty affecting mostly young adults during their most productive years



Healthy Lungs – Jo Dietrich & Katharine Honey

Mixed/canvas
(100 x 300 cm)

'By my bedside a young woman is laughing
How can I come to her alone, without my body?
I can't leave it, I must not touch her
My touch, like the touch of death, spreads disarray'

The Body and Us (Poem) – Antun Branko Šimić

Jo Dietrich

Denmark

Jo Dietrich studied at the Department of Media, Cognition and Communication, Faculty of Humanities, University of Copenhagen, two years at the Department of Psychology, University College London and Institute of Education, London. In connection with this, she visited UNESCO in Paris.

Her main skills are coordination, administration and networking; participatory and qualitative research and evaluation skills; and knowledge of international development policies and development goals, particularly about gender education and HIV/AIDS.

She has worked with HIV/AIDS in developing countries and in Denmark for a number of years, and she now runs the project 'Art against AIDS' for the Danish AIDS Foundation, in which she facilitates art workshops for women with different cultural backgrounds. She does contemporary abstract paintings.

Katharine Honey

Katharine Honey studied painting and stained glass at the Wimbledon School of Art, then taught at the Institute of Education before training with the jewellery designer Miye Matsukata in the United States. Later she returned to London and further consolidated her studies at Sir John Cass and more recently at Morley College. Working mostly in silver and using semi-precious stones, her work emphasizes colour and texture within an abstract form. While some of her work is commissioned, she is now focusing on producing a more coherent range of jewellery, with occasional designs in a limited edition. Her work has been exhibited at the Oxford Gallery; Crafts at Rangers House, English Heritage, Greenwich Tourist Office, Cockpits Arts Morley College, Saltgrass Gallery, and the Greenwich Open Studios.

CHAPTER 24

Preclinical and Clinical Development of New Tuberculosis Vaccines: Regulatory Requirements and the Transition to Phase I and Beyond

Kenneth Barry Walker

International Regulatory Framework: A Brief Overview

Since the days of Edward Jenner and the origins of vaccine technology, concerns over safety and quality of vaccines have been raised, along with recognition of the public health benefits accruing from a well-structured vaccination programme. With increasing use of vaccines, and a number of severe adverse events consequent to production and quality failures during the first part of the twentieth century, there came a realization that it was important to have an independent evaluation of medicinal products before they are released for public health uses, this conclusion being arrived at different times in different regions of the world. Consequently, a framework for regulating vaccines, pharmaceuticals, and biological agents for human and veterinary use was developed. This framework has been extended and refined over the years and is now a major legal force in many countries, regulating and ensuring the

quality, safety, and efficacy of vaccines in particular, but also including other biological therapeutics, pharmaceuticals, and extending to some aspects of herbal medicines and the cosmetics industry.

In the USA these requirements for the efficacy and safety of medicines resulted in the formation of one of the first of these regulatory authorities—what is now known as the Food and Drug Administration (FDA). The FDA is a scientific, regulatory, and public health agency whose jurisdiction encompasses most food products (other than meat and poultry), human and animal drugs, therapeutic agents of biological origin, medical devices, radiation-emitting products for consumer, medical, and occupational use, cosmetics, and animal feed.

The modern era of the FDA dates to 1906 with the passage of the Federal Food and Drugs Act (1) adding regulatory functions to the agency's scientific mission. After a series of changes in name and remit, the agency was transferred to the Department of Health, Education, and Welfare (HEW). By 1968 the FDA became part of the Public Health Service within HEW, and in 1980 the education function was removed from HEW to create the Department of Health and Human Services, FDA's current home. Within the FDA, the Centre for Biologics Evaluation and Research (CBER) (recently renamed Vaccines, Blood & Biologics (2)) regulates biological products including vaccines and has the delegated authority for issuing manufacturing and marketing licences for this class of products. The structure of CBER is complex and it has a number of offices and divisions with separate delegated responsibilities (see Appendix 24.1). FDA scientists evaluate applications for new human drugs and biologics, complex medical devices, food and colour additives, infant formulas, and animal drugs. The FDA investigates and inspects manufacturing facilities, ensuring compliance with the relevant manufacturing requirements. Biological products intended for veterinary use are regulated under a separate law, the Virus, Serum, and Toxin Act, which is administered by the US Department of Agriculture (3).

The period of the 1960s and 1970s saw, for most countries—whether or not they had initiated pharmaceutical product registration controls earlier—a rapid increase in laws, regulations, and guidelines for reporting and evaluating the data on safety, quality, and efficacy of new medicinal products and most countries now have regulatory authorities (RAs) carrying out similar functions, or at least with similar responsibilities. For example, Japan, Australia, China, and Canada all have related national regulatory authorities (NRAs) performing similar functions regulating vaccines and biologics for human use. An extensive list of national regulatory authorities can be located at <http://www.fda.gov/oia/agencies.htm>.

The increasing internationalization of the pharmaceutical/biological industry was eventually recognized as being somewhat at odds with the reality that

registration of medicines remained a national responsibility. Although different regulatory systems were based on the same fundamental obligations to ensure the quality, safety, and efficacy of new medicinal products, the detailed technical requirements between these different authorities had diverged over time. It was recognized that there was a case to be made for action to rationalize and harmonize regulation between nations in order to ensure there is a minimum of delay in making safe and efficacious new treatments available to patients in need.

The first stages of this process in Europe were undertaken in 1965 with the implementation of the first stages of a legislative framework for handling European pharmaceuticals (EC directive 65/65/EEC) (4, 5). Over the next decade, further developments ensued which culminated in the formation of the Committee for Proprietary Medicinal Products (CPMP), the predecessor of the existing EMEA (6). Problems and complications remained and a major review of market authorization within the EU was undertaken in the late 1980s which resulted in the implementation of what was termed the 'Future Systems' package which was launched in January 1995. This provided the legislative framework for the formation of the European Medicines Agency (EMA) which acts as a pan European regulatory authority, accompanied by the relevant nation-state competent authorities. The EMA has its head office at Canary Wharf, London. The EMA is the EU's body responsible for coordinating scientific resources put at its disposal by individual member states for the evaluation, supervision, and pharmaco-vigilance of medicinal products. Each member state has pooled its sovereignty for the authorization of medicines through the establishment of a 'centralized procedure' for the licensing of pharmaceuticals for human use. Accompanying this centralized procedure is the 'mutual recognition' process which provides for nationally issued market authorizations to be accepted and recognized across Europe. Interestingly, with the centralized procedure, the EMA itself is not empowered to issue market authorizations, that is, to license products. Licensure or market authorization is actually provided by the European Commission, based upon the recommendations and opinions provided through the EMA (6, 7).

The EMA provides the relevant competent authorities of the member states of the EU the scientific advice on any question relating to the evaluation of the quality, safety, and efficacy of medicinal products for human or veterinary use, referred to it in accordance with the provisions of EU legislation relating to medicinal products. They also have responsibility for ensuring facilities and manufacturing processes are within specification and appropriate processes and procedures are being followed. This includes on-site inspections of manufacturing facilities and also audit of current good manufacturing practices (CGMP) documentation and compliance records. The most recently amended legislation for the European Medicines Agency is (EC) No 726/2004 (6, 8).

Harmonization of regulatory requirements was pioneered by the European Community (EC) in the 1980s, as the EC (now the European Union) moved towards the development of a single market for pharmaceuticals. The success achieved in Europe with the EMEA demonstrated that international harmonization was a realistic possibility and an International Committee for Harmonization (ICH) was proposed to help undertake rationalization and harmonization of the regulatory framework. The WHO Conference of Drug Regulatory Authorities (ICDRA), in Paris in 1989, crystallized these discussions and specific plans for action began to materialize. Soon afterwards, the authorities approached the International Federation of Pharmaceutical Manufacturers and Associations (IFPMA) to discuss a joint regulatory–industry initiative on international harmonization and, subsequently, the ICH was formed, with the first meeting being held in Brussels in April 1990 (9).

The long-term goal of developing a harmonized format has led to the creation of the ICH Guideline M4: the Common Technical Document (CTD) (9, 10). This was agreed as an important target for the first phase of ICH activities and was designed to remove redundancy and duplication in the differing national product development and review processes. The outcome would be that a single set of data, collated into a common format dossier, could be generated to demonstrate the quality, safety, and efficacy of a new medicinal product and that dataset would be in a format and structure that could be formally recognized across international boundaries. The CTD provides a harmonized format and defines the content for new product applications. The agreed upon implementation date for the CTD, in three regions (Europe, Japan, and the United States) was July 2003 (9, 10).

To further streamline the registration and licensure processes, the Electronic Common Technical Document (eCTD) (11) was developed subsequently through the ICH process. This electronic document format allows for the electronic submission of the CTD direct from applicant to regulatory body and provides a harmonized technical solution to implementing the CTD electronically. The eCTD has been implemented across the ICH partner and observer regions and has greatly facilitated and improved the market authorization process.

From the point of view of vaccine developers, the development of harmonized procedures for licensure and medicinal product regulation has provided more clear pathways to licensure. However, there are numerous scientific, technical, and administrative hurdles remaining. Many of these are generic issues that will apply across many product types; however, there will always be a range of product-specific issues that relate to the nature, contents, formulation, or proposed use of the candidate vaccine. A range of these issues and the potential regulatory issues specifically associated with the development of TB vaccines are discussed further below.

Generic Issues Relating to Tuberculosis Vaccines

Preclinical Development

Preclinical development is the earliest part of the product life cycle, undertaken once some experimental proof of principle has been established and a decision has been made to take the vaccine strategy forward to develop into a product. This will involve the development of preliminary protocols for product characterization, with the view to possibly develop these protocols into manufacturing release specifications in due course. There will also be extensive studies both *in vitro* and *in vivo* to establish safety, efficacy, and potency, often using relevant animal models, where possible. The accumulation of safety data is of critical importance during this stage of the development process, along with the development of possible potency assays and an attempt to determine relevant surrogate markers of vaccine induced protection (12–16).

The nature of the data required to progress the development of the product at this stage of the product life cycle is driven by the product developer—and investors, if present. The evidence collected in these studies will need to be sufficiently clear and robust to provide confidence to the developer and possible investor that further development is appropriate. Although regulatory interest at this point is minimal, however, due recognition should be given to the nature of the data that will need to be presented eventually to RAs for approval for clinical trial. These data may also eventually constitute part of the documentation supplied for licensure (in the eCTD). Broadly speaking, compliance with the requirements and structure of the CTD at the earliest stages will facilitate the development process and will aid in ensuring the appropriate data are captured and presented in an appropriate form. It is, therefore, prudent to initiate early discussions with RAs or engage those experienced in the regulatory processes of the country concerned at the earliest opportunity. There are a wide range of guidance documents available through this portal at the EMEA website <http://www.emea.europa.eu/index/indexh1.htm>.

CGMP and GLP Compliance

Although formal compliance with GLP and CGMP standards is not required during early stages of *preclinical* development, the establishment of laboratory regimes and processes that are broadly compliant with GLP will help ensure that the required information is traceable and that the product history and development is properly recorded. For example, dedicated laboratory notebooks and a process of regular signing off (weekly or monthly) of the laboratory book by a responsible senior scientist is helpful for tracing development

history and product handling, transfer, and modifications. In the case of live attenuated or genetically modified vaccine candidates, a clear history of the parent organism and all changes to it should be collated at the earliest possible instance. Laboratory seed stocks should be stored appropriately and preserved at different physical locations, as early as possible. This should be with a high level of documentation and traceability being maintained (17–19). Again, an understanding of the general requirements of the CTD and what RAs might wish, as a consequence to see, will be helpful to the developer (18, 20, 21).

Potency Assays: Animal Models

It is likely to be the case that potency assays (in vitro or in vivo), or the relevance of specific animal models, in determining go/no go decision points in preclinical development will need to be decided and rationalized by the developer and financial sponsors. There are no universally accepted animal models, or surrogates of potency, as yet with TB vaccines and so the responsibility lies with the vaccine developer(s) to establish the appropriate and justifiable criteria to progress their product.

Decisions on animal models and surrogate markers can have a significant impact on downstream development; for example, if an in vitro assay for a surrogate marker indicative of immunogenicity leading to protection has an elegant scientific rationalization, but it is not functionally possible to qualify or validate the assay to the satisfaction of RAs, then data from that assay may be very difficult to justify assays in clinical trials or for manufacturing processes. The RAs will critically review the assay systems being used or proposed and, therefore, a robust and defensible data set is critical to winning the RAs approval to proceed. It has been known that in later stages of product development, specific assays have not been accepted as capable of assessing product equivalence in the face of material changes to product production parameters and so may pose significant and unexpected development difficulties (22–26).

Aspects of Contracting Out Work

There are a number of aspects of the preclinical development process that may be amenable to being contracted out (e.g. preclinical toxicology leading to Phase I studies, or specific physicochemical or analytical procedures) and appropriate recognition of the importance of relevant expertise, specific capabilities, and GLP accreditation status should be taken into account during the decision-making processes leading to the award of a contract-testing arrangement.

Clearly there are number of highly complex, product-specific issues associated with the preclinical phase of product development, many of a scientific nature; however, sight should not be lost of the underlying purpose of the preclinical development process—to bring a scientific concept through early development

to the point of entry to Phase I clinical trial. In achieving success in this process it has become evident that the scientist/developer can learn many lessons from the processes developed by trial and error in manufacturing and pharmaceutical industries, many of these lessons and outcomes being unrecognized by the scientific researcher. To bridge this gap, access to relevant expertise through scientific and project management advisory groups and the development of a specific and targeted clinical development plan should be undertaken as early as possible. Allocation of funds to engage expert consultants at key points and the recruitment of a project manager with relevant product development expertise can be critical to reducing the number of potentially delaying decisions made during the preclinical stages leading to manufacture of a product for clinical trial (27).

Preclinical to Clinical Development Plan and CMOs

The details of any clinical development plan will be product dependent and tailored around the nature of the product, its final target product profile (dose, route, indication, and target population), and pragmatic manufacturing issues that will inevitably arise during product development. Appropriate key stages and implementation points will need to be identified and strategies for alternative outcomes identified, in line with good project management practice (e.g. PRINCE2 is one of a number of accredited project management frameworks), to aid the smooth development of the product. A potentially critical step in the clinical development plan will be the identification and selection of a suitable manufacturing facility (Contract Manufacturing Organization or CMO), this possibly being associated with the identification of an industrial partner if not already engaged in the project.

It should not need to be stated at this point that all products destined for clinical trial in the countries within the umbrella of the ICH (and almost everywhere else) are required to be manufactured to CGMP standards in an appropriately accredited facility (17–19, 21). The CMO is a critical choice—their expertise, background in pharmaceutical or biological manufacturing, and level of formal accreditation (GLP, CGMP) will be major factors in the decision-making processes by the product developers (27). The development of open and clear channels of communication between all partners as well as clear criteria for product development and manufacture will need to be established at the earliest stages, along with the mutual recognition that flexibility and product change are an inherent part of development of an Investigational Medicinal Product (IMP).

Once a CMO has been chosen and engaged there will begin the often complex task of product-specific technology transfer, ensuring standard operating procedures are in place, that appropriate training has been implemented and

recorded, product material transfer has been achieved, and any issues arising from scale-up are being addressed. Product specifications will need to be developed along with manufacturing release criteria. Frequent meetings between all levels involved are important in this process and it needs to be recognized that it will often be the individuals at the sharp end of assay development and technology transfer that will require the most support and attention to detail to ensure smooth transfer. The developer will need to recognize their responsibilities and to review progress, audit the CMO, or engage experts who can audit the CMO, at regular agreed intervals or critical points.

The CMO, in conjunction with the developer, project manager, and any other relevant parties will need to establish the programme details for manufacturing to Phase I, including specifications for batch consistency, and stability, and to identify areas where contract outsourcing may be necessary. There are many documents available through the ICH, the EMEA, and the FDA that describe both in outline and in detail the requirements during this stage of the product development cycle (17–21, 28).

Vaccine Issues: Generic and Specific

Generic Issues

Vaccines against TB have undergone rapid development and deployment into early clinical trials in the last decade. The details of these vaccines vary enormously, with fundamental differences in scientific approach, formulation, and application. Current investigational vaccines against TB range across:

- Recombinant proteins, potentially with a variety of adjuvants
- Non-living vectored vaccines, using a range of vectors (this includes Nucleic Acid vaccines)
- Live attenuated MTB and derivatives
- Recombinant live bacterial vaccines based upon BCG, this includes other live bacterial vectors
- Bacterial extracts and purified bacterial components

Each of these approaches and technology platforms has with it specific regulatory issues and questions which need to be considered; however, there remain common themes based upon the framework of regulatory requirements that are broadly applicable to all the TB vaccines—issues of quality, safety, and efficacy. There is extensive literature available along with guidance documents from regulatory agency websites (EMEA, ICH, and FDA) and so only some specific issues will be highlighted here.

Quality

Quality of production and the effects the target product profile (dose, route, proposed clinical indication, and target population) has upon production issues are central to all vaccines. Consequently, early identification of the production methodology, scale-up issues, and establishing the product specifications are critical components in ensuring the quality of the final product. It is a general requirement that agents for human use (including early clinical trial: Phase I/II) are manufactured to CGMP specifications, in an approved facility with all the requisite controls, documentation, and infrastructure. There are numerous Guidelines (29–37) that provide focus to the development of a good quality profile that will fit well within the structure of CTD or eCTD—see the ICH, EMEA, or FDA websites for more details.

Safety

Safety issues associated with novel immunological products going first-in-man have become higher profile since the TGN1412 incident in 2006. A number of recent guidelines outlining Phase I clinical trial strategies for so called ‘high risk’ products have been published and, although at present none of the proposed TB vaccine candidates fall into this ‘high risk’ category, familiarization with the guidelines would be appropriate for all developers considering first-in-man studies (12, 14, 38–41). For all vaccines there are issues of inappropriate immune responsiveness and local tolerability and these can be of particular interest with TB vaccines given the complex nature of the immune response to TB both in health and disease.

TB pathology and disease resolution both involve similar immune pathways, with regulation of the cytokine network being a key component modulating both disease progression and disease clearance or disease prevention (after immunization). This has long been recognized as a risk factor in immunization strategies and much scientific discussion and experimental work has been devoted to clarifying the theoretical versus the practical risks of adverse immune outcomes after vaccination with TB vaccines (42–46). The TST is an example of the basis for concerns of inappropriate immune responses. In this test, often used as a diagnostic for TB or as an indication of BCG ‘take’ after BCG vaccination, a small amount of a purified protein extract of MTB is injected intradermally into the forearm. This can result in a pronounced and lasting reactive immune response at the site of vaccination, characterized by inflammation, redness, cell infiltration, and can last for up to a week. The test outcome is determined by measuring the diameter of the induration or swelling across the injection site 24 and 48 hours after injection. In naïve individuals, without mycobacterial exposure, the response is very small or non-existent; however, those infected with TB or with a history of BCG vaccination can respond profoundly to the

antigenic stimulation with a large uncomfortable swelling that can persist for some time. As a diagnostic test for infection, and as a potential indicator of protection against infection after vaccination, the TST has a chequered history; however, the immune basis of the response should not be ignored.

This often profound immune response to injected mycobacterial antigens subsequent to infection, or if there is a history of BCG vaccination, leads to the possibility of hyper-responsiveness in vaccine recipients and, indeed, the possibility of exacerbation of disease in those carrying dormant or subclinical TB. This last concern has been a point of contention for some years with a number of robust publications demonstrating a risk in animal models of TB with some classes of vaccines (47–50). To date in clinical trials, vaccination with TB vaccines, even in higher risk groups (e.g. PPD+, TB exposed), has not seen a worrying increased level of local adverse events or systemic adverse events, and so the perceived risk appears to be diminishing as experience is acquired with these products. The matter, however, remains live and is a necessary and significant consideration in planning clinical trials of TB vaccines, particularly Phase I safety in at risk populations (48).

Efficacy

The efficacy of a vaccine in a given population can only be determined pragmatically from the outcome of, firstly, a Phase III efficacy trial and, subsequently, post market surveillance of efficacy in the field. However, in the preclinical data package, proof of principle of efficacy, through relevant protection studies in animal models, will be required (12, 13, 51–53). These data from animal studies will support both product development go/no go decisions as well as supporting the application for Phase I trial. But it must be remembered that even in the face of a successful Phase III efficacy study, the dossier for market authorization (eCTD) will require extensive and scientifically sound data and the preclinical studies that support the proposed efficacy of the product (52, 54).

Specific Issues Related to Vaccine Classes

Recombinant Protein and Adjuvant Combinations

New TB vaccines based upon recombinant antigens and existing or novel adjuvants have been developed and some are already in clinical trial. Recombinant fusion proteins and multiepitope proteins have been developed and have been shown to have an effective preclinical profile with respect to immunogenicity and safety in animal models. Recently, specific fusion proteins have been combined with novel adjuvants (e.g. IC31 or CFA01 to mention just two) and been shown to be safe and immunogenic in both animals and humans. Current regulatory

requirements mean that for entry to Phase I studies, both the fusion protein, as well as the adjuvant itself, must have a full CGMP quality and manufacturer dossier for preparation and characterization, with specifications for release and relevant safety data. In addition, the final formulation, adjuvant and protein combination in liquid or freeze dried form, must also have a full quality and characterization dossier complying with CGMP quality requirements. The format and content structure of the eCTD can provide much assistance on the nature and detail of the data required in the dossier, but be in no doubt that the information required will need to be comprehensive (55).

The main issues that arise with this family of new TB vaccines primarily revolve around the adjuvant, its interactions with regard to the final formulation and safety issues with the adjuvant alone, and in combination. Where the adjuvant proposed is novel, a comprehensive dossier on manufacturing, safety, quality, and immune interactions will need to be compiled (55). Recombinant fusion proteins, although often being amenable to a good characterization package, will however, require information on the nature of the responses elicited by the fusion protein as compared to native proteins, the benefits, and the immune interactions occurring as a consequence of the nature of the construct. This is often evidenced in terms of antigen-specific immune responses of a relevant nature, with the current emphasis being mostly directed towards measurement of IFN- γ production by antigen-specific T cells by a variety of technology platforms (56, 57).

Non-Replicating Viral-Vectored Vaccines (including DNA vaccines)

Live non-replicating vectored vaccines, including MVA and Adenovirus (e.g. Ad35 amongst a number of others), can be considered a subset of live vaccines, given that for manufacturing they must replicate and assemble appropriately and are grown on various cell substrates. However, a vital difference from other live vaccines is that they are designed to be replication-deficient in human cells and, therefore, have an extremely limited replicative cycle once administered. The safety implications are greater than those found with recombinant protein vaccines; however, the safety record of the viral vectors being used or considered for use in TB vaccines is usually robust and, for example, MVA is considered safe to administer even to immune-compromised individuals (58–60).

The issues of vaccine characterization and manufacturing quality remain as generic issues; however, the details are often notably more complicated for this class of product than that found in recombinant proteins. This complexity arises as a consequence of the active component (the viral-vectored construct) requiring growth and replication in a cell substrate with the complexities of characterization and quality assessment that this entails. The ICH, EMEA, and FDA all have guidelines on issues related to manufacturing of biologicals

for human use (including vaccines) in live cell substrates and these guidelines should be consulted early on during product development (34, 61).

As this class of product, being a 'live' viral vector expressing specific recombinant proteins, is classified as a genetically modified organism (GMO) this poses some additional concerns from both the RAs and from other government agencies such as those responsible for agriculture and environment. In the preclinical development stages, laboratory containment itself and compliance with what in the UK is known as 'contained use regulations' is often sufficient with little more than standard GMO/laboratory precautions being needed. However, experience to date has shown there will be a requirement for a detailed environmental impact assessment as well as a risk and mitigation strategy for first-in-man studies (62, 63). The requirements for these impact statements vary widely depending on the country and the national sensitivities. There is a large political/social acceptability component in these concerns and so the scientific basis for the GMO issues may be less well founded than some of the product safety and quality concerns, this in no way suggesting that they need to be dealt with any less rigorously (62–65). It should also be noted that national processes for dealing with GMO/environmental issues are often completely separate procedures with their own time line and due consideration should be given to these factors.

Live Attenuated Vaccines

At present, the most advanced live vaccine candidates are based on recombinant BCG or rationally attenuated MTB. It is testimony to the changing landscape of TB vaccine development that live vaccines have a place in the forefront of TB vaccine platforms. As recently as 20 years ago the considerations of live TB vaccines based upon either recombinant BCG or the possibility of rationally attenuating pathogenic MTB were mostly negative. The regulatory hurdles were considerable and no clear pathways through these hurdles were available; also the technology platforms for manipulating the organism were still under development (62, 64). However, as our understanding of the biology of the organism has grown, along with the understanding of human immunology, the possibility that live TB vaccines could fill a vital role of affecting a viable vaccine approach has become clearer.

In parallel with this, the regulatory experience with recombinant organisms and novel vaccine technologies has broadened, breaking down some of the barriers to the possibility of live TB vaccines. Indeed, within the last five years, a recombinant BCG candidate vaccine has entered Phase 1 clinical trial and others are in advanced preclinical development (43, 44, 66). Nevertheless, the hurdles that face live vaccines that have entered clinical trial and future live vaccines remain considerable.

To clarify some of these issues with live vaccines (particularly those based on mycobacteria) a meeting was convened in 2005 in Geneva to directly approach the highest priority concerns and attempt to draw together a consensus from world experts on the answers to some of the pressing questions that faced developers and regulators assessing these vaccines. The outcome of this meeting was the Geneva Consensus on Live Mycobacterial Vaccines (48) and within it a number of clear recommendations were agreed that could clarify and facilitate development of this class of vaccine. In particular, the absence of antibiotic or enzyme markers was strongly urged; the requirement for two independent mutations in the case of attenuated MTB vaccines was noted. In principle, either of the two mutations should be able to retain the attenuated phenotype in the unlikely event one mutation was lost. Issues of quality, characterization, and stability were addressed; some recommendations for master seed, working seed, and lot production were also noted. It has been noted that the WHO recommendations for BCG (and the European Pharmacopea (EuPh)) have specifications for the number of passages from master seed to product that would require proof of stability (8 passages for EuPh and 12 passages for the WHO recommendations). The techniques employed to demonstrate genetic stability in the case of new TB vaccines can utilize modern molecular techniques, for example, targeted polymerase chain reaction (PCR) or blotting technology.

The safety issues associated with live mycobacterial vaccines against TB are in principle the same; however, because these products are live and/or attenuated from pathogenic organisms, clearly the level of proof will be somewhat higher. The Geneva consensus document noted the EuPh and WHO recommendations for BCG safety studies and duly noted that this new class of vaccines would require additional proof of safety through targeted animal studies. Comparative survival and dissemination studies in normal and immune-compromised mice were recognized as being particularly useful. Guinea pigs, as a highly susceptible species, also should contribute to the potential portfolio of safety studies (48).

Bacterial Extracts and Purified Proteins

Many of the basic issues relating to this class of product apply also to the recombinant protein approach. However, in addition, the clearest complications with this class of vaccine are the characterization and manufacturing issues. Characterization can often become a point of concern as the preclinical development plan proceeds and the need for product specifications becomes more pressing. Some of the extracts proposed as potential vaccines are either relatively crude, or purified but complex materials. In both instances, characterization of the product can be difficult as the active component in the crude mix, or the complex nature of the molecule (e.g. multiple glycosylation

sites) can make the setting of specification for production a problem. Related to this will be the ability to demonstrate consistency of manufacture as extraction protocols are noted for their variability and, unless the process development is rigorous and closely monitored, the ability to consistently characterize the material may be in jeopardy. The presence of 'contaminating' material in these products is always an issue but can be markedly alleviated with proper and rigorous process development, along with a sophisticated process monitoring. Efforts should be made early in preclinical development to address these issues as without appropriate management the overall development plan may be put at risk. The ICH, EMEA, and FDA websites have much guidance relating to quality and should be consulted (a few of these are noted herewith (30–34, 67–70)).

Clinical Trials

Having successfully navigated the rocky waters of preclinical development and consideration is being given to the Phase I trial, there are some product specific issues that need to be considered. Fortunately, the vast majority of the requirements for a Phase I trial are clearly laid out in a variety of documents available on the Internet or can be acquired through the services of companies that specialize in designing, managing, and running clinical trials (40, 71–74).

The main specific issues that arise from trials using TB vaccines are the potential for additional and possibly specialist monitoring for safety issues, the possible use of novel adjuvants, and issues relating to the use of live organisms. These may require additional resources, the possibility of additional arms to the study to provide information on novel adjuvants, and management of environmental concerns for live vaccines (48).

There are many documents available relating to Good Clinical Practice (GCP) in clinical trials, clinical trial design, and monitoring as well as the relative responsibilities of the parties involved. The clear enunciation of the primary end point (safety) and its measurement and details of attainment are required as well as a clear understanding of the implications of any secondary end points that may be included (immunogenicity is quite often a secondary end point) (40, 74–76). Immunogenicity in the context of TB trials is an area of intense discussion at the moment. There are a limited number of well-characterized assay systems available and they are used in clinical trials for a number of vaccine trials for other diseases (HIV and malaria) including TB. They are based on ELISPOT technology, intracellular flow cytometry, and whole blood antigen stimulation. The read-outs for all these assays are currently focused on

IFN- γ production (56, 77–84). Due caution on the weight given to the output from these assays must be applied and the difficulties in developing these assays further from laboratory and investigative tools to fully qualified assays should not be underestimated. It must be remembered that validated assays are required for application in Phase III trials (23, 26, 37, 85).

The basic Phase I study usually uses healthy normal volunteers; however, there are a number of other considerations where TB vaccines are concerned. The final target population may well be neonates or young children and so due consideration should be given to the overall strategy that will need to be applied to attaining the requisite Phase I safety data in the target population. In a similar vein, there are clear ‘increased risk’ groups as has been noted earlier—in particular those that may have prior BCG vaccination, those exposed to TB, or those subclinically infected. Again, due consideration is required on the overall trial strategy that will provide the required safety data. A further ‘at risk group’ is clearly those that are immune-compromised, where there are two considerations: firstly, vaccine efficacy in the face of a compromised immune system and; secondly, the safety issues of live vaccines in immune-compromised individuals.

GMO Issues and Clinical Trials

The issues of GMO status for a range of the vaccine technologies being proposed has implications for the management of clinical trials, not the least for subsequent field trials in endemic countries and potential licensure. For the early Phase I trials, the indications are that the main concerns are focused on containing spread (shedding), providing data on persistence and dissemination, and providing reasoned scientific arguments concerning environmental risks. These have, to date, been sufficient to allow Phase I studies to commence. There is potential for these issues to reduce in importance as experience is obtained in Phase I trials; however, the risks associated with the need to transfer Phase I studies to endemic countries should not be underestimated. Each nation has its own risk management model for GMO-related issues and some have a very high bar for the nature of the argument required to persuade the authorities to permit release in Phase I or II trials. Early and serious consideration should be given to this matter, taking into account the longer term strategic clinical development plan.

Conclusion

The last 20 years has seen remarkable advances in our understanding of TB, both as an organism and as a disease. Many of these advances have fed directly

Appendix 24.1 CBER Structure (from CBER Website (2))

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into the development of specific vaccine platforms or specific potential vaccine products. From a position where there were no recognized possible vaccine candidates, there are now active clinical trials underway, a number of new products in late preclinical development, and many exciting new approaches being developed. The probability of seeing a replacement vaccine for BCG or a successful booster for BCG in the foreseeable future is now appreciably improved and it is now generally accepted in the field that it is a matter of when, not if.

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