Technologies, standards and norms

Standards for biological products

The 64th meeting of the WHO Expert Committee on Biological Standardization was held in Geneva from 21-25 October 2013. The large number of participants in attendance reflected the strong need expressed by Member States to WHO for support to appropriately regulate biological products.

The international standards established by the Committee are designed to be common global standards that promote regulatory convergence between countries. This is accomplished when the WHO standards are adapted into many national regulations, and when they are used by the WHO Prequalification Programme as the compliance standard for UN procurement. During the meeting, three new written standards were adopted that define regulatory expectations for biotherapeutic products, for adjuvanted vaccines, and for typhoid conjugate vaccines.

Biotherapeutic products
Developments in molecular genetics and nucleic acid chemistry have opened up new avenues for the production of medicines. Genes encoding natural biologically active proteins can be identified, modified and transferred from one organism to another in order to obtain highly efficient synthesis of their products. New recombinant DNA (rDNA)-derived biological medicines are now produced using a range of different expression systems such as bacteria, yeast, transformed cell lines of mammalian origin (including human origin), insect and plant cells, as well as transgenic animals and plants. rDNA technology is also used to produce biologically active proteins that do not exist in nature, such as chimeric, humanized or fully human monoclonal antibodies, or antibody-related proteins or other engineered biological medicines such as fusion proteins.

Together these technologies have enabled the production of large quantities of medicinal products that are difficult to prepare from natural sources or were previously unavailable. Nevertheless, it is still not possible to fully predict the biological properties and clinical performance of these macromolecules on the basis of their physicochemical characteristics alone. In addition, they are produced in biological systems which are known to be inherently variable – with important consequences for the safety and efficacy of the resulting product. Therefore, before such biologicals are introduced into routine clinical use it must be ensured that their quality is consistent from lot to lot. This is achieved by developing robust manufacturing processes on the basis of process understanding and characterization, with appropriate in-process controls. Process understanding and consistency are critical since slight changes can occasionally have a major unwanted impact, for example on immunogenicity, with potentially serious safety implications.

The new guidelines on biotherapeutic products (1) are intended to provide
national regulatory authorities (NRAs) and manufacturers with guidance on the quality, safety and efficacy of rDNA-derived biotherapeutics) and intended for use in humans. The guidelines are based on experience gained over three decades in this technically demanding field. Part A sets out updated guidelines for the manufacture and quality control of rDNA-derived biotherapeutics, including consideration of the effects of manufacturing changes and of devices used in the delivery of the product and on its stability. Part B provides guidelines on nonclinical evaluation, while Part C provides guidance on clinical evaluation. Product-specific vaccine-related recommendations and guidelines are available elsewhere\(^1\), as are additional considerations for similar biotherapeutic products (2).

**Adjuvanted vaccines**

The second global standard adopted by the Expert Committee provides guidance to NRAs and manufacturers on the nonclinical and initial clinical evaluation of vaccine adjuvants and adjuvanted vaccines (3) by outlining the international regulatory expectations in this area.

Over the past decades, new approaches have been devised to develop and deliver vaccine antigens. Some of these antigens are weakly immunogenic and require the presence of adjuvants to induce or enhance an adequate immune response. Vaccines with aluminium-based adjuvants have been used extensively in immunization programmes worldwide, and a significant body of safety information has accumulated for them. As science and technology have advanced, vaccines containing adjuvants other than aluminium-containing compounds (e.g. human papillomavirus and hepatitis B vaccines) have been authorized for use in many countries, and a number of vaccines with novel adjuvants are currently under development, such as vaccines against human immunodeficiency virus (HIV), malaria and tuberculosis, as well as new-generation vaccines against influenza and other diseases.

However, the development and evaluation of adjuvanted vaccines present regulatory challenges. Vaccine manufacturers and regulators have questions about the type of information and extent of data that would be required before adjuvanted vaccines can proceed to clinical trials and eventually be authorized for use. Existing WHO guidelines on nonclinical evaluation of vaccines (4) give valuable general guidance but provide limited information specifically related to new adjuvants and adjuvanted vaccines.

The new guideline provides updated and more extensive guidance on the nonclinical and preclinical testing of adjuvants and adjuvanted vaccines. It should allow manufacturers and regulators to proceed on the critical path towards licensure of adjuvanted vaccines that will help to control some diseases with important global public health impact.

**Typhoid conjugate vaccines**

The third set of guidelines adopted by the Committee are intended to assist NRAs in evaluating the scientific issues connected with the quality, safety and efficacy of typhoid conjugate vaccines that use Vi polysaccharide covalently linked to a carrier protein (5). The available guidelines for Vi polysaccharide typhoid vaccine (6) and for live, attenuated Ty21a vaccines (7) are not applicable to this type of typhoid vaccines, which have carrier proteins such as diphtheria toxoid (DT), tetanus toxoid (TT), recombinant *Pseudomonas aeruginosa* exoprotein A (rEPA), the
nontoxic mutated form of diphtheria toxin – for example, cross-reactive material 197 (CRM197) – or another suitable protein.

The evidence gathered thus far indicates that typhoid conjugate vaccines may have several advantages over unconjugated Vi polysaccharide vaccines, including: (i) greater efficacy and effectiveness; (ii) longer persistence of immunity; (iii) immunogenicity across all age groups, including infants and toddlers aged younger than 2 years; (iv) perhaps some degree of herd immunity; and (v) induction of immune memory with initial dosing, leading to anamnestic responses to a subsequent dose or doses.

The guidelines are based on experience gained during the development of experimental typhoid conjugate vaccines as well as relevant information obtained from the evidence for other types of bacterial polysaccharide–protein conjugate vaccines, such as Haemophilus influenzae type b (Hib), and meningococcal and pneumococcal conjugate vaccines. Part A of the guidelines sets out guidance on manufacturing and quality control, while Part B addresses the nonclinical evaluation of these vaccines and Part C addresses their clinical evaluation. Part D provides guidance for NRAs.

Reference preparations
The provision of internationally accepted biological reference preparations is an important normative activity of WHO. These global measurement standards enable the efficacy, quality, purity and safety of very many biological products to be stated in a common language worldwide. Thus, biological reference preparations support:
• Biological and immunological assays for the quality control of a wide range of biologicals – therapeutics, blood-derived products, vaccines and immunological products of traditional types – as well as those derived from modern biotechnological approaches.
• Standardization of materials and approaches used in medical diagnostics such as diagnosing disease, monitoring therapy, blood safety, and public health applications (such as monitoring immune status, screening for disease or susceptibility) or otherwise characterizing biological material from individuals.
• Development, evaluation, standardization and control of products by industry, by regulatory authorities, and also in biological research in academia and scientific organizations. They play a vital role in facilitating the transfer of laboratory science into worldwide clinical practice and the development of safe and effective biologicals.

These reference preparations are provided to Member States to calibrate national, or regional, quality control materials for biological medicines and regulated diagnostic tests. The latter, for example, enable quantitative limits to be expressed in regulations in standardized units of measurement.

New reference preparations
Twelve new international biological reference preparations were adopted by the Expert Committee (see Table 1) and have been added to the catalogue of WHO biological reference preparations for blood products and related substances2.

Discontinued reference preparations
The Committee agreed to discontinue the following reference preparations which were considered no longer fit for purpose:
• anti-echinococcus serum (code number ECHS 75.1106);
• anti-C complete blood typing serum (code number W1004 84.1424);

2 http://www.who.int/bloodproducts/catalogue/Bloo2014.pdf
The Committee endorsed a number of new projects. The timely development of new reference materials and standards is critically important to harness scientific developments for new biologicals. At the same time, the active management of the existing inventory of reference preparations requires a carefully planned programme of work to replace established materials before the stock of containers, which comprises the standard, is exhausted. The Committee agreed to the initiation of the following new projects on reference preparations for vaccines and related materials:

- anti-E complete blood-typing serum, human (code number W1005 83.1424).

### New projects

The Committee endorsed a number of new projects. The timely development of new reference materials and standards is critically important to harness scientific developments for new biologicals. At the same time, the active management of the existing inventory of reference preparations requires a carefully planned programme of work to replace established materials before the stock of containers, which comprises the standard, is exhausted. The Committee agreed to the initiation of the following new projects on reference preparations for vaccines and related materials:

<table>
<thead>
<tr>
<th>Preparation</th>
<th>Activity</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pegylated Granulocyte Colony Stimulating Factor</td>
<td>10 000 IU per ampoule</td>
<td>First WHO International Standard (IS)</td>
</tr>
<tr>
<td>Tumor Necrosis Factor alpha, recombinant, for bioassay</td>
<td>43 000 IU per ampoule</td>
<td>Third WHO IS</td>
</tr>
</tbody>
</table>

#### In vitro diagnostic device reagents:

<table>
<thead>
<tr>
<th>Preparation</th>
<th>Activity</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Antibodies to Hepatitis B virus “e” antigen (anti-HBe) **</td>
<td>120 IU/mL</td>
<td>First IIS</td>
</tr>
<tr>
<td>Hepatitis A virus RNA for NAT-based assays *</td>
<td>54 000 IU/mL</td>
<td>Second IS</td>
</tr>
<tr>
<td>Hepatitis B virus “e” antigen (HBeAg) **</td>
<td>100 IU/mL</td>
<td>First IS</td>
</tr>
<tr>
<td>Hepatitis D virus RNA for NAT-based assays **</td>
<td>575 000 IU/mL</td>
<td>First IS</td>
</tr>
<tr>
<td>HIV-1 Circulating Recombinant Forms RNA for NAT-based assays *</td>
<td>Ten panel members consisting of CRFs and other variants. No unitage assigned</td>
<td>First International Reference Panel</td>
</tr>
<tr>
<td>Human Serum IgE *</td>
<td>13 500 IU/mL</td>
<td>Third IS</td>
</tr>
<tr>
<td>Mycoplasma DNA for NAT-based assays designed for generic mycoplasma detection **</td>
<td>200 000 IU/mL</td>
<td>First IS</td>
</tr>
<tr>
<td>Parvovirus B19 DNA for NAT-based assays *</td>
<td>1 410 000 IU/mL</td>
<td>Third IS</td>
</tr>
</tbody>
</table>

#### Vaccines:

<table>
<thead>
<tr>
<th>Preparation</th>
<th>Activity</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trivalent inactivated polio vaccine, for D antigen assay</td>
<td>277 DU/ml for poliovirus type 1, 65 DU/ml for poliovirus type 2, 248 DU/ml for poliovirus type 3</td>
<td>Third IS</td>
</tr>
</tbody>
</table>

Notes:

- Vaccines and related substances, cytokines, growth factors and biotherapeutics other than blood products, and in vitro diagnostic device reagents identified by * are held and distributed by the National Institute for Biological Standards and Control, Potters Bar, Herts, EN6 3QG, England.
- In vitro diagnostic device reagents identified by ** are held and distributed by the Paul-Ehrlich-Institut, 63225 Langen, Germany.
Technologies, standards and norms

- diphtheria toxoid for flocculation test (3rd IS);
- meningococcal Serogroup A polysaccharide (1st IS);
- typhoid Vi polysaccharide (1st IS);
- high and low mutant virus reference preparations for MAPREC assay of poliovirus type 2 (2nd IS);
- antiserum to Respiratory Syncytial Virus (1st IS).

Regarding the proposal for generic approval of standards and reference panels for cancer diagnostics, the Committee felt that this represents a large commitment that needs clarification from WHO in regard to the Committee’s focus and resources.

No requests were received to initiate new projects for cytokines, growth factors and endocrinological substances or for antibiotics.

The Committee agreed to the initiation of the following new projects on reference preparations on blood products and in vitro diagnostic devices:
- replacement of Hepatitis C Virus RNA for NAT assays (5th IS);
- Anti-Cytomegalovirus IgG;
- Malaria (P. falciparum) antibody reference panel;
- high and low titre anti-A and anti-B in serum/plasma;
- anti-Rubella Immunoglobulin;
- replacement of Anti-Tetanus Immunoglobulin (2nd IS);
- assignment of FIX antigen value to 4th IS/5thFIX plasma/concentrate;
- replacement of Ancrod (2nd IS);
- replacement of Streptokinase (4th IS).

References