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EXPERT COMMITTEE ON BIOLOGICAL STANDARDIZATION

Fifth Report

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EXPERT COMMITTEE ON BIOLOGICAL STANDARDIZATION

Fifth Session

Geneva, 3-8 December 1951

Members :

Dr. J. H. Gaddum, Professor of Pharmacology, Edinburgh University, Edinburgh, United Kingdom

Professeur E. Grasset, Directeur de l'Institut d'Hygiène ; Professeur de Bactériologie et d'Hygiène à l'Université de Genève, Geneva, Switzerland (*Vice-Chairman*)

Dr. N. K. Jerne, Acting Chief, Department of Biological Standards, Statens Seruminstitut, Copenhagen, Denmark

Dr. A. A. Miles, Director, Department of Biological Standards, National Institute for Medical Research (Medical Research Council), London, United Kingdom (*Chairman*)

Dr. C. A. Morrell, Director, Food and Drugs Divisions, Department of National Health and Welfare, Ottawa, Canada

Dr. J. Ørskov, Director, Statens Seruminstitut, Copenhagen, Denmark

Dr. W. G. Workman, Chief, Biologics Control Laboratory, National Microbiological Institute, National Institutes of Health (US Public Health Service), Bethesda, Md., USA

Dr. P. M. Wagle, Director, Haffkine Institute, Bombay, India, was unable to attend.

Advisers :

*Sir Thomas Dalling, Chief Veterinary Officer, Animal Health Division, Ministry of Agriculture and Fisheries, London, United Kingdom (Representative of the Joint WHO/FAO Expert Group on Zoonoses)

Dr. W. L. M. Perry, National Institute for Medical Research (Medical Research Council), London, United Kingdom

Consultants :

*Dr. H. H. Green, Ministry of Agriculture and Fisheries Veterinary Laboratory, New Haw, Weybridge, Surrey, United Kingdom

*Dr. A. W. Stableforth, Director, Ministry of Agriculture and Fisheries Veterinary Laboratory, New Haw, Weybridge, Surrey, United Kingdom

Secretariat :

Dr. W. Aeg. Timmerman, Director, Division of Therapeutic Substances, WHO (*Secretary*)

Dr. M. M. Kaplan, Chief Veterinary Officer, Division of Epidemiological Services, WHO

Dr. R. Pollitzer, Consultant on Cholera, Division of Epidemiological Services, WHO

The report on the fifth session of this committee was originally issued in mimeographed form as document WHO/BS/136, 18 December 1951.

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* Attended one day of the session to assist the committee with regard to special subjects.

EXPERT COMMITTEE ON BIOLOGICAL STANDARDIZATION

Fifth Report [1]

The fifth session of the Expert Committee on Biological Standardization was held in Geneva from 3 to 8 December 1951.

The Deputy Director-General welcomed the experts, explained certain changes in the regulations for expert committees, and emphasized that the experts attended meetings in their capacity as individuals and not as representatives of countries. He had noted that the committee had an important agenda before it, which included studies on the standardization of diagnostic procedures, a function of WHO laid down in Article 2(*t*) of the Constitution. In discussing the problems of veterinary standards, the committee had taken the initiative in proving that WHO recognized no difference between veterinary and human problems in medicine. WHO was grateful to the experts for giving up their time in order to attend this session.

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[1] The Executive Board, at its ninth session, adopted the following resolution:

I. The Executive Board

1. NOTES the report of the Expert Committee on Biological Standardization on its fifth session;

2. THANKS the members of the committee for their work, and

3. AUTHORIZES publication of the report.

II. Considering that direct contact between the international centres in Copenhagen and London and the national control centres in Member States has considerable advantages with regard to the functioning of national centres;

Considering that regional control centres might lead to unnecessary complications and prejudice the efficient working of the national control centres,

The Executive Board

RECOMMENDS that no regional control centres be established.

III. Noting that considerable progress has been made in the establishment of international standard preparations for diphtheria toxoids;

Considering that it is not therefore necessary to refer this matter to an expert committee,

The Executive Board

REQUESTS the Director-General not to convene in 1952 the expert committee on diphtheria toxoid standardization for which provision was made in the budget.

(Resolution EB9.R84, *Off. Rec. World Hlth Org.* **40**, 30)

1. Cholera [2]

The committee noted the progress made by the Statens Seruminstitut, Copenhagen, (1) in the establishment of dried standard preparations of the cholera vibrios to be used in the preparation of diagnostic antisera, and (2) in the collaborative assay of freeze-dried cholera vaccines in terms of the provisional reference vaccine.

The committee noted the recommendation made by the Expert Committee on Cholera that a single strain of cholera-susceptible mice should be made available for use in the assay of cholera vaccines.[3] The committee recommended that breeding-stock of such a strain should be made available from a single source.

In answer to an observation of the Expert Committee on Cholera, that freeze-dried antiserum might be unsuitable as a standard because antisera sometimes lose potency during freeze-drying,[3] the committee pointed out that the potency of a standard should always be assigned to the freeze-dried preparation after reconstitution, and that loss of potency during freeze-drying could, therefore, be ignored.

2. Pertussis Vaccine [4]

The committee noted the progress made by the Statens Seruminstitut in its investigation of the proposed standard preparation of pertussis vaccine, and of the best methods of assay.

3. Diphtheria Toxoid [5]

The committee established as the international standard for diphtheria toxoid, plain, the provisional reference preparation of diphtheria toxoid, plain, which is a relatively highly purified toxoid and which had proved to be of the same order of potency as current therapeutic preparations in various countries. The committee also considered the relative merits of crude and partially-purified plain toxoids as standards, and decided that the new standard would serve adequately for the assay of diphtheria toxoids

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[2] Statens Seruminstitut, Copenhagen, unpublished working document WHO/BS/130

[3] *World Hlth Org. techn. Rep. Ser.* 1952, **52**, 12

[4] Statens Seruminstitut, Copenhagen, unpublished working document WHO/BS/123

[5] Statens Seruminstitut, Copenhagen, unpublished working document WHO/BS/113

of all degrees of purity. The committee noted that the slopes of the dosage-response lines for plain diphtheria toxoids tended to become flatter as the purity increased, but considered that in practice such differences in slopes would not seriously affect the validity of the assays.

The committee authorized the Statens Seruminstitut to proceed with the establishment of an international reference preparation of diphtheria toxoid, adsorbed. The Statens Seruminstitut was authorized to obtain a sample of freeze-dried adsorbed toxoid similar to that at present in use at the Paul Ehrlich Institute, Frankfurt-on-Main, Germany, and to institute a collaborative examination of the material, including accelerated degradation tests.

The committee deferred assignment of unitage to both the international standard for diphtheria toxoid, plain, and the proposed international reference preparation of diphtheria toxoid, adsorbed, and asked the Statens Seruminstitut to collect data on the relation between the immunizing potency of plain and adsorbed toxoids in man, with a view to assigning to the international standard for diphtheria toxoid, plain, a unitage approximately equivalent to that of the proposed international reference preparation of diphtheria toxoid, adsorbed.

The committee re-affirmed the principle that the size of a new international unit should be assigned so as to avoid, as far as possible, any change in the size of existing well-established units. In cases where the international unit was made equivalent to an existing unit, the committee would expect either that the designation of the existing unit should be replaced by the term " international unit " or at least that the designation should include a definition of its relation to the international unit.

In this connexion the committee decided that in assigning a unitage to the proposed international reference preparation of diphtheria toxoid, adsorbed, every effort would be made to relate it to the " Schutzeinheit " of the German standard for diphtheria toxoid, held by the Paul Ehrlich Institute, Frankfurt-on-Main.

4. Tetanus Toxoid [6]

The committee established the international standard for tetanus toxoid. The standard preparation had proved to have an immunizing potency similar to current therapeutic preparations of tetanus toxoid in various countries. Since there was evidence that the slopes of the dosage-response

[6] Statens Seruminstitut, Copenhagen, unpublished working documents WHO/BS/125, WHO/BS/125 Add.1, WHO/BS/125 Add.2

lines for plain and adsorbed toxoids were closely similar, a single standard could at present be used for the assay of both plain and adsorbed preparations of tetanus toxoid. The committee defined the international unit of tetanus toxoid as the immunizing activity of 0.03 mg of the international standard for tetanus toxoid. In accordance with the principle re-affirmed in section 3 (page 5) of this report, this unit was made approximately equivalent to the " Schutzeinheit" of the standard tetanus toxoid held by the Paul Ehrlich Institute, Frankfurt-on-Main.

5. Streptococcus Antitoxin

The committee noted the progress made in the establishment of an international standard for streptococcus antitoxin and the different magnitudes of national unit potencies already established. There was evidence of heterogeneity of streptococcus antitoxins, similar to that observed in tetanus antitoxins. The committee decided to defer the establishment of this standard and authorized the National Institute for Medical Research, London, to continue the investigation of the heterogeneity and to obtain opinions about the unit potency to be assigned to the standard.

6. Enteric and Rickettsial Diagnostic Antisera

The committee noted that the preparations of *Salmonella* diagnostic antisera were ready for collaborative study of their suitability as international standards. The progress made in preparing *Proteus* diagnostic antisera was also noted.

7. Tuberculin

7.1 *Mammalian PPD* [7]

The committee established the batch of purified protein derivative (PPD) of tuberculin, prepared by Dr. Florence Seibert of the Henry Phipps Institute, Philadelphia, Pa., USA, from a human strain of the tubercle bacillus, as the international standard for purified protein derivative of mammalian tuberculin. The unitage of this standard will be assigned as soon as possible.

Although the committee intends that the standard shall serve for the assay of PPD tuberculins used in the diagnosis of both bovine and human infections, it recognized that for certain purposes the assay of bovine PPD

[7] Green, H. H., unpublished working document WHO/BS/127

in terms of a human PPD standard might prove to be insufficiently specific. The committee, therefore, decided to include in the designation of the standard a statement that the international standard preparation is a specimen of PPD from a human strain of the tubercle bacillus.

7.2 *Avian PPD* [8]

The committee decided to establish an international standard preparation of PPD of avian tuberculin and accepted the offer by the Veterinary Laboratory of the Ministry of Agriculture and Fisheries (England and Wales), Weybridge, Surrey, United Kingdom, of the existing standard of that Ministry as the proposed international standard preparation. It authorized the Weybridge Veterinary Laboratory, in consultation with the appropriate committees of the Food and Agriculture Organization of the United Nations (FAO) and the International Office of Epizootics, to proceed with a collaborative examination of its suitability as an international standard preparation and of the unitage to be assigned to it.

The committee also noted that the Weybridge Veterinary Laboratory is prepared to make available to interested workers the strain (D4) of avian tubercle bacillus which was used in the manufacture of the proposed international standard preparation.

7.3 *Assay of tuberculin* [9]

The committee noted certain improved methods of assay of tuberculin which make possible the calculation of fiducial limits of error of the estimated potency of tuberculin from the internal evidence of the assay.

8. Organic Arsenical Substances

8.1 *Oxophenarsine* [10]

On the basis of the results of a collaborative examination of the existing joint Canadian-British standard for oxophenarsine, the committee decided to establish it as the international standard for oxophenarsine. The committee authorized the National Institute for Medical Research to obtain opinions from interested workers about the necessity for this standard in the light of the definitive chemical constitution of preparations of oxophenarsine.

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[8] Green, H. H., unpublished working document WHO/BS/126

[9] Long, D. A., Miles, A. A. & Perry, W. L. M., unpublished working document WHO/BS/120

[10] Department of Biological Standards, National Institute for Medical Research, London, unpublished working document WHO/BS/133

8.2 *Melaminyl trypanocides* [11]

The committee considered the request of the International Scientific Committee for Trypanosomiasis Research that standard preparations should be provided for melaminyl trypanocides.

The committee decided to establish international reference preparations of two of these substances, namely Melarsen (the sodium salt of *p*-(2:4-diamino-*s*-triazinyl-6-)-aminophenylarsonic acid) and its polymerized antimonial analogue, designated " MSb ", and authorized the National Institute for Medical Research to obtain preparations of these two substances and to institute a collaborative study of their suitability as international reference preparations.

9. Dimercaprol

The committee noted the progress made by the National Institute for Medical Research in obtaining opinions of interested workers about the suitability of the British standard for dimercaprol as an international standard.

10. Diagnosis of Syphilis

10.1 *Syphilitic sera*

The committee noted the progress made by the Subcommittee on Serology and Laboratory Aspects of the Expert Committee on Venereal Infections and Treponematoses in the establishment of a group of reference syphilitic sera for the calibration of various serological tests for syphilis. The results of a pilot experiment indicated that freeze-dried sera were suitable for the preservation of specific and non-specific reactivity. The committee recommended to the Subcommittee on Serology and Laboratory Aspects that further tests be made of the stability of preparations at temperatures above 37°C.

10.2 *Cardiolipin and lecithin* [12]

The committee established provisional international reference preparations of cardiolipin and lecithin. It authorized the Statens Seruminstitut to consult with the New York State Department of Health, Albany, N.Y., USA, about the suitability for this purpose of the preparations already

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[11] Lourie, E. M., unpublished working document WHO/BS/134

[12] Pangborn, M. C., Maltaner, F., Tompkins, V. N., Beecher, T., Thompson, W. R. & Flynn, M. R. (1951) *Cardiolipin antigens*, Geneva (*World Health Organization : Monograph Series*, No. 6); unpublished working document WHO/Pharm/Mon/465 Rev. 2