

World Health Organization

Technical Report Series

No. 28

EXPERT COMMITTEE ON RABIES

Report on the First Session

Geneva, 17-22 April 1950

	Page
1. Introduction	4
2. Improvement of types of vaccine in current use	5
3. New types of vaccine	6
4. Standardized potency-tests for vaccines	6
5. Virus strains and rabies vaccine production	7
6. Hyperimmune antirabies serum	8
7. Field trial of antirabies serum	8
8. Biological test for confirmatory diagnosis of rabies	9
9. Local treatment of animal-bite wounds	9
10. Indications for vaccine treatment	9
11. Control of rabies in animals	11
12. Regional meetings	13
13. International conference	14
14. Field demonstration of rabies control in dogs, involving the use of vaccine	14
15. Collection and evaluation of rabies statistics	14
16. Indications for future lines of research	15
Annex 1. Notes on method of preparation of ultraviolet irradiated vaccine ("UV" vaccine)	18
Annex 2. Technique of preparation of avianized rabies vaccine	18
Annex 3. Potency mouse test (Habel) (fixed virus)	19
Annex 4. Modification of potency mouse test (fixed virus)	20
Annex 5. Standard challenge virus for potency mouse tests	20
Annex 6. Potency guinea-pig test (Koprowski) (street virus)	21
Annex 7. Preparation of hyperimmune antirabies serum	22
Annex 8. Feasibility of a trial of hyperimmune serum in man: cases of rabid-wolf bites in Iran	23
Annex 9. Field trial of antirabies hyperimmune serum in human beings exposed to rabid-wolf bites	24
Annex 10. Suggested technique of biological test for confirmatory diagnosis of rabies	25
Annex 11. Preparation of rabies vaccine with removal of paralysis-producing factor	26
Annex 12. Guinea-pig test for paralysis-producing factor	26

WORLD HEALTH ORGANIZATION

PALAIS DES NATIONS

GENEVA

NOVEMBER 1950

SECOND IMPRESSION, DECEMBER 1951

THIRD IMPRESSION, AUGUST 1953

EXPERT COMMITTEE ON RABIES

First Session

Members :

- Dr M. L. Ahuja, Director, Central Research Institute, Kasauli, East Punjab, India
- Dr I. A. Galloway, Director, Virus Research Institute, Pirbright, Surrey, United Kingdom (*Chairman*)
- Dr K. Habel, Chief, Laboratory of Infectious Diseases, Microbiological Institute, National Institutes of Health (US Public Health Service), Bethesda, Md., USA
- Dr H. N. Johnson, Laboratories of the International Health Division, Rockefeller Foundation, New York City, N.Y., USA
- Dr H. Koprowski, Assistant Director, Section of Viral and Rickettsial Research, Lederle Laboratories, Pearl River, N.Y., USA (*Rapporteur*)
- Dr P. Lépine, Chef du Service des Ultra-Virus, Institut Pasteur, Paris, France (*Vice-Chairman*)

Co-opted Member :

- Dr M. Baltazard, Directeur de l'Institut Pasteur de l'Iran, Teheran, Iran

Secretaries :

- Dr M. M. Kaplan, Veterinary Officer, Epidemiological Studies Section, WHO
- Dr C. Klimt, Medical Officer, Epidemiological Studies Section, WHO

The report on the first session of this committee was originally issued in mimeographed form as document WHO/Rabies/17, 24 April 1950.

EXPERT COMMITTEE ON RABIES

Report on the First Session¹

The problem of rabies was considered by the First World Health Assembly,² whereupon the Executive Board at its second session instructed the Secretariat to submit a questionnaire to experts on rabies.³

The Second World Health Assembly recommended that an expert committee be convened in 1950 to consider the answers to the questionnaire as prepared by the Secretariat late in 1949, to make recommendations as to the desirability of holding an international rabies conference, and to initiate research in which antirabies institutes and authorities would

¹ In accordance with the instructions of the Executive Board at its fifth session, an ad hoc committee examined this report and, acting on behalf of the Board, adopted the following resolution :

The ad hoc committee of the Executive Board

1. NOTES the report of the Expert Committee on Rabies on its first session ;
2. AUTHORIZES its publication ;
3. REQUESTS the Director-General
 - (1) to arrange, as approved by the Executive Board at its fifth session, the undertaking of a field trial using hyperimmune serum-vaccine as a preventive measure for rabies in man, and a field demonstration on the control of rabies in dogs using a new avianized vaccine ;
 - (2) to co-ordinate the exchange of virus strains for the production and testing of rabies vaccines ;
 - (3) to arrange regional meetings of appropriate authorities from neighbouring and nearby countries where rabies is a problem so that concerted attacks on this disease will be possible, and
 - (4) to encourage, wherever possible, research on problems in rabies requiring clarification, as indicated in section 16 of the report of the Expert Committee on Rabies ;
4. DECIDES to defer a decision on convening an international conference on rabies pending examination of the results of the afore-mentioned field trials at the second session of the Expert Committee on Rabies ;
5. TRANSMITS the present report to the Third World Health Assembly, and
6. POINTS OUT that recommendations of expert committees which concern WHO policy and operations remain recommendations unless and until they are implemented by the Executive Board or the World Health Assembly in adopting and putting into action the annual programme of WHO.

(*Off. Rec. World Hlth Org.* 28, Annex 2)

² *Off. Rec. World Hlth Org.* 13, 144

³ *Off. Rec. World Hlth Org.* 14, 21

be invited to participate.⁴ Furthermore, the Executive Board at its fifth session passed the following resolution :

“ The Executive Board,

On condition that the Expert Committee on Rabies at its first session early in 1950 so advises,

AUTHORIZES the undertaking of WHO-sponsored field trials with a minimum of delay in (a) the use of a new hyperimmune serum-vaccine prophylactic measure against rabies in human beings, and (b) the use of a new egg-adapted virus vaccine in dogs for the control and eradication of rabies.”⁵

The Expert Committee on Rabies held its first session in Geneva from 17 to 22 April 1950. Dr I. A. Galloway was elected Chairman, Dr P. Lépine, Vice-Chairman, and Dr H. Koprowski, Rapporteur.

1. Introduction

The committee, before submitting its observations and recommendations, considers it opportune to summarize briefly the most recent important advances in the study of rabies.

The introduction of the use of the mouse as an experimental animal has provided a means of securing a more accurate quantitative determination of virus potency and the elaboration of a more reliable and useful method of confirmation of histopathological findings (Negri bodies) by application of a biological test. Again, as a result of the application of what are now commonly referred to as “potency mouse tests”, it has been possible to establish a better means of measuring antigenic content of vaccines and their immunizing or protective value. This has led to a very great improvement in the quality of vaccines produced by different institutes and commercial firms.

Moreover, it was the introduction of the use of the mouse for detecting virus and making quantitative determinations that led to the development of a new and very effective vaccine, the ultraviolet light irradiated virus vaccine, or what is now freely termed “UV” vaccine.

Another step forward has been the demonstration of the possibility of propagating the virus in chick embryos. This led to the development of another prophylactic agent of great value and promise, avianized virus vaccine.

⁴ *Off. Rec. World Hlth Org.* **18**, 139 ; **21**, 182

⁵ *Off. Rec. World Hlth Org.* **25**, 8

There have been extensive observations on the immunization of dogs with products resulting from laboratory investigations to improve methods of preparing vaccines and of determining their potency or protective value. The tests that have been made in this direction have been carried out more satisfactorily than in the past, and it has been possible to attain a more reliable assessment of the efficacy of vaccines. Very encouraging results have been obtained; and, as improved methods of vaccination are being introduced, there is a trend towards a wider application of vaccination of dogs to supplement other methods of control of the disease.

Re-evaluation of the use of hyperimmune antirabies serum has indicated from an experimental standpoint its great promise, when combined with the use of vaccine, for the prevention of rabies in man.

The recognition of important vectors of rabies virus other than dogs, cats, wolves, jackals, and foxes, e.g., the *Viverridae* (mongoose, meercat, and genet) in the Union of South Africa, and the vampire bats in South and Central American States, has suggested the need for further ecological studies since, arising out of the observations on vampire bats as vectors, the existence of asymptomatic carriers has been demonstrated.

More recently, efforts have been concentrated in another important direction: the development of techniques for the study of the paralysis-producing properties of the virus-infected brain tissue from which antirabies vaccines are commonly prepared, and the devising of ways of removing these by appropriate means.

As a result of its deliberations, the committee is in a position to submit the following observations and recommendations.

2. Improvement of Types of Vaccine in Current Use

Through the use of mice in potency tests for virus antigenicity, the importance of various steps in the production of the types of vaccine already established has been evaluated. Such factors as the high titre of the original brain suspension, the higher concentration of the emulsion of tissue, the smaller amount of inactivating chemical, the determination of the exact time required to inactivate the virus, the influence of the temperature of inactivation — all have been shown to improve the immunizing potencies of certain vaccines. This system of checking the influence of all steps in the process of vaccine production by experimental, quantitative determination of their effects, and its general application to specific types of vaccines in current use, are strongly recommended to all vaccine-producing laboratories.

3. New Types of Vaccine

3.1 *Ultraviolet irradiated vaccine ("UV" vaccine)*

The introduction of irradiation of rabies virus suspensions by ultraviolet light, under strictly controlled conditions, has led to the development of a new method of inactivating virus, while preserving its antigenicity or immunizing properties. The uniformly high potency of such properly irradiated vaccines, their ability to withstand storage under suitable conditions, and the adequate demonstration of the safety of their use as vaccines in man and dog, justify the recommendation that UV vaccines be added to the list of acceptable vaccines (see Annex 1).

3.2 *Avianized vaccine*

This vaccine contains active virus (Flury strain) modified and attenuated by passage in developing chick embryos. It has been shown that this virus propagated in chick embryos is devoid of pathogenicity for dogs when injected parenterally, i.e., by intramuscular (masseter) and subcutaneous inoculation. No cases of postvaccinal paralysis have been observed. In vaccination experiments, dogs which received a single injection of avianized vaccine proved to be resistant to the inoculation into the masseter muscles of rabies "street" virus (dog salivary gland). Recent observations have shown that all dogs immunized with avianized vaccine were resistant, one year later, to infection with rabies street virus (see Annex 2).

The stage was reached when it was felt that the testing of this type of vaccine under appropriate conditions of natural, as distinct from experimental, infection was indicated. Field tests are already in progress, with satisfactory results so far, as to the safety of the vaccine under conditions prevailing in the USA.⁶

4. Standardized Potency-Tests for Vaccines

The committee stresses the great value of the potency mouse test for estimating the immunizing value of vaccines, and the necessity for its wider application as a routine procedure (see Annex 3).

While it is recognized that this now well-known potency mouse test (Habel) is of the greatest value, consideration is being given to a slight modification of this type of test in the direction of employing varying

⁶ In the list of recommendations for future research, mention is made of other new approaches to the development of improved prophylactic agents (see section 16, page 15).

dilutions or doses of vaccine against an appropriate standard dose of the "fixed" virus for challenge (see Annex 4).

Furthermore, it must be appreciated that in special cases, and for the purpose of accumulating comparative data, another type of potency test is available and has proved of value in giving significantly consistent results. In this test a challenging dose of a suitable strain of street virus is given by the intramuscular route, and the test animals may be either hamsters or guinea-pigs (see Annex 6).

The committee is strongly of the opinion that it is desirable to set up some mechanism⁷ whereby (1) it would be possible for vaccine-producing centres to procure a sample of a suitable strain of fixed rabies virus for challenge purposes in the potency mouse tests, and (2) it would be possible, periodically, for vaccine-producing centres to have the potency of samples of some batches of their vaccine checked. This would increase their confidence in their methods of production.

5. Virus Strains and Rabies Vaccine Production⁸

General experience throughout the world has indicated that many of the characteristics of street virus strains differ with the animal source and the geographical origin. Such properties as invasiveness, incubation period, symptoms produced, relative susceptibility of various species of experimental animals, may show wide variation, but qualitatively all strains of rabies virus seem to have a common antigenic constitution.

This is quite important since a choice must be made as to which strain of virus will be used as the source of antigen for vaccine production. At the present time, evidence purporting to show the superiority of a fixed strain derived from local street virus would seem to have been based on experiments lacking quantitative treatment. In the present state of our knowledge, therefore, there would appear to be no scientific basis for replacing the Pasteur strain of fixed virus by local strains for the production of the usual type of rabies vaccine.

However, some interesting findings on the variation of antigenic structure of classical fixed virus strains have been reported. Substrains of the

⁷ The committee feels that because of the many variables which characterize viruses, the establishment of a "standard" rabies virus and of a "standard" rabies vaccine could not be conceived having regard to the exacting requirements of the WHO Expert Committee on Biological Standardization for the types of product with which they are accustomed to deal. It would appear, therefore, that that body could not be expected to assume responsibility for the organization of the service which the Expert Committee on Rabies visualizes.

⁸ For resolution adopted by the ad hoc committee of the Executive Board, see section 3, paragraph (2) of footnote 1, page 3.

Pasteur fixed virus, maintained by animal passage in various laboratories, have apparently changed in their properties, including their antigenic pattern. It has been found by cross-immunity and cross-neutralization tests that some substrains give good protection against challenge by most of the others, whereas other substrains give little such protection. Likewise, from the standpoint of choice of the strain to be used as the challenge in the potency mouse test, it was found that whereas one substrain was able to show vaccines made from a number of other substrains to be highly immunizing, another used as challenge was able to break through the immunity produced by all substrains. For this reason the particular substrain selected for use on the standard challenge virus was one of intermediate invasiveness (see also Annex 5).

It would seem pertinent in view of these findings to encourage the checking of all production strains by cross-immunity tests with other available substrains of fixed virus, in order to obtain information on the breadth of their antigenic constitution.

6. Hyperimmune Antirabies Serum ⁹

For many years the possible prophylactic use of rabies-immune serum has been recognized, and numerous experiments have pointed to its practical potentialities. More recent experimental work employing quantitative procedures has definitely shown the superiority of hyperimmune serum, especially when combined with a course of vaccine, over the use of vaccine alone after exposure to peripherally introduced street virus. Practical means of producing this serum in large animals such as sheep and the concentration of the specific antibody are available (see Annex 7). The use of highly potent hyperimmune serum preceding a course of vaccine gives promise of saving most of those cases of human rabies in which a short incubation time does not allow a sufficiently long period for the development of active immunity. In view of these experimental findings, the committee feels that at the present time serum combined with vaccine offers the best promise as a means of preventing rabies after severe exposure, and strongly recommends the setting-up of a field trial in human beings thus exposed.

7. Field Trial of Antirabies Serum

In order to implement the recommendations of the preceding section, consideration was given to the possibility of having such a field trial carried out under optimal conditions. After a considerable amount of discussion,

⁹ For resolution adopted by the ad hoc committee of the Executive Board, see section 3, paragraph (1) of footnote 1, page 3.

it was decided that such a trial should take place in Iran. Dr Baltazard, Director of the Institut Pasteur, Teheran, would be responsible for carrying out this trial. (A description of the relevant local conditions will be found in Annex 8, and the detailed conditions as laid down for this trial in Annex 9.)

The committee requested that the appropriate secretary of the committee should be responsible for following the trial, checking the data, and passing the results to the members of the committee. It was agreed that such data will be regarded as confidential, and no use will be made of them before their evaluation and release at the next session of the committee.

A limited amount of serum has been made available for this field trial. If further amounts should become available, a field trial should be envisaged in India. Appropriate data for this purpose are to be collected by WHO.

8. Biological Test for Confirmatory Diagnosis of Rabies

The confirmatory intracerebral inoculation of mice or hamsters for the presence of street virus in instances of questionable diagnosis on pathological grounds is definitely recommended. The mouse is the animal of choice but, where not easily available, hamsters may be substituted because of their high susceptibility to street virus infection (see Annex 10).

9. Local Treatment of Animal-Bite Wounds

The committee recommends the immediate treatment of bite wounds inflicted by rabid animals, or animals suspected of being rabid, by thorough cleansing with soap or detergent solution. Such treatment does not preclude the subsequent application of agents for the suppression of bacterial contamination, such as antibiotics.

Later, as information is obtained on the protective value of antirabies hyperimmune serum, consideration should be given to its use for local treatment by infiltration of the tissue about the site of exposure.

10. Indications for Vaccine Treatment

The committee considered the variation in indications for vaccine treatment at present used throughout the world. There was general agreement along the lines set forth in table I, which has been formulated with a view to reducing to a minimum the number of persons subjected to treatment unnecessarily.

TABLE I. INDICATIONS FOR VACCINE TREATMENT

Nature of exposure	Condition of biting animal		Decision as to vaccine treatment at time of possible exposure
	At time of exposure	During observation period of 10 days	
I. No lesions ; indirect contact only	healthy or rabid	healthy or rabid	none
II. Licks : (1) unabraded skin (2) abraded skin or mucosa	(a) healthy	healthy or rabid healthy	none none
	(b) healthy	clinically suspicious or proven rabid	start treatment at appearance of first suspicious signs
	(c) suspicious	healthy	start treatment immediately ; stop treatment if animal remains normal for 3 days
	(d) animal rabid, escaped, killed, or unknown		start treatment immediately
III. Bites	(a) healthy	healthy	no treatment, except if bites are multiple, or face, head or neck bites ; then treat as in III (c)
	(b) healthy	clinically suspicious or proven rabid	start treatment at appearance of first suspicious signs
	(c) suspicious	healthy	start treatment immediately ; stop treatment if animal remains normal for 3 days
	(d) animal rabid, escaped, killed, or unknown ; or any bites by jackal, wolf, fox, or other wild animal		start treatment immediately

Note : Bites on the head, neck, and shoulders, deep multiple wounds, and those inflicted by wild animals involve a greater degree of risk, and patients should be treated accordingly.

Fairly often a situation arises in which a person previously exposed to infection and treated with vaccine is re-exposed to infection with rabies. The question as to whether treatment should be re-instituted and, if so, on what basis, must be answered. It is recommended that, if this situation arises within three months of the first course of vaccine, no further treatment is necessary unless the second exposure is of a severe type. If the interval is between three and six months, two reinforcing doses of vaccine, one week apart, are indicated, whereas if more than a six-months' interval has elapsed the treatment should be on the same basis as if it were an original exposure.

Occasionally, marked allergy to rabies vaccine manifested by angio-neurotic oedema, fever, adenopathy, shock, etc., is encountered. This may be during the course of immunization or, more often, following the administration of the first dose to a person who has previously received rabies vaccine. It is suggested that this difficulty may be circumvented

by a change to vaccine made from the brain tissue of another species of animal (i.e., from rabbit-brain vaccine to sheep-brain vaccine).

11. Control of Rabies in Animals

It is recognized that rabies exists in two epizootiological forms : (1) a widely disseminated disease propagated principally in dogs, predominately in urban regions ; and (2) a more localized disease of wild animals, particularly in wolves, foxes, jackals, vampire bats, and mongoose.

The application of known effective measures for the elimination of rabies from the dog population constitutes the most challenging problem at this time in that this animal is the principal source of human infection.

The committee has considered the answers to the questionnaire concerning the control of rabies in animals, and notes the general agreement on the main principles to be applied in rabies control. On the other hand, there are wide differences of opinion expressed with regard to fundamental details, such as the length of quarantine periods, and the testing and application of veterinary vaccines. The committee wishes to record, therefore, their recommendations on these various problems, with the realization that the application of the various measures proposed will have to be adapted to local conditions.

The committee recognizes the distinct value of periodical, compulsory, prophylactic vaccination of dogs against rabies, and recommends its use in areas in which the disease is enzootic.

During the past ten years carefully controlled experiments have demonstrated the value of a single dose of either living or inactivated virus vaccine, repeated annually. From a practical point of view, the efficacy of this procedure has been corroborated by the extensive and successful field use of inactivated virus vaccines in the USA, and the continued success with living attenuated virus vaccines in Hungary.

The committee recommends that vaccines be subjected to adequate tests for potency. It is of the opinion that the presence of live virus in vaccines is not of itself sufficient proof of antigenicity, and recommends the use of the potency mouse test (Habel) for both living and inactivated virus vaccines (see Annex 3). The committee appreciates, however, that the promising new avianized live virus vaccine (Flury strain) appears to present a different problem in potency testing which has now been adequately met (see Annex 6). This vaccine has proved to be highly antigenic in tests in guinea-pigs and dogs.

The committee recommends that where feasible a biting animal should be kept under observation for a period of ten days. If the animal shows

no signs of illness during this period, it can safely be assumed that the animal was non-infective at the time of biting.

The committee further recommends that during an outbreak of rabies, if general restrictive measures alone are depended upon in an involved area, dogs should be restrained (leashing, secure confinement) for a minimum period of 90 days from the date of the last known case of rabies. Where, in addition to restrictive measures, vaccination of dogs is carried out, the period of restraint may be reduced to 30 days after vaccination. The restraint of domestic cats is not feasible.

The committee recommends that dogs and cats bitten by a rabid animal should be destroyed. If the owner is not willing to destroy the exposed animal, the following alternatives are recommended :

- (a) strict isolation of the animal in a kennel for a period of 6 months ;
- (b) if no previous vaccination has been given within a period of 12 months, vaccination and confinement in a kennel for 3 months ;
- (c) if the animal has been previously vaccinated within 12 months, revaccination and restraint (leashing, secure confinement) for 30 days.

As regards domestic livestock exposed to rabies by bite, it is recognized that these animals will probably not propagate rabies. Exposed animals can be slaughtered for meat purposes within one week of the bite, or after six months. It should be noted that the greatest risk in dealing with exposed livestock is the danger encountered by people handling the live animal or the carcass, and not through the consumption of meat or milk from infected animals. Because of the lack of controlled experiments, the committee does not feel it possible to make any specific recommendation with respect to the vaccination of large animals following exposure.

The committee recognizes that countries now free of rabies should continue either to prohibit the importation of dogs and cats, or subject them to a prolonged period of quarantine, preferably six months, at the port of entry. In the case of countries with extensive land borders, and where rabies is already present in domestic or wild animals, it is recognized that such strict quarantine measures are impracticable. There can be no objections to the importation of dogs from countries free of rabies provided they have been isolated en route. Dogs originating in infected countries should be vaccinated within 12 months before departure, and revaccinated as soon as possible after arrival, by whatever procedure is practical in a particular area.

These recommendations are made in consideration of the varied conditions encountered throughout the world, and should not be construed as discouraging more stringent measures, such as quarantine periods upon entry, with (preferably) or without vaccination.

Experience has shown that the efficient organization of a rabies-control programme in an infected area is best accomplished by means of a central authority headed by a public-health officer, preferably a veterinarian, who has full executive power and who devotes his full time to this work. A system of weekly reports of rabies cases should be instituted to enable the officer to keep abreast of the problem. He should enlist the support of all local groups directly or indirectly concerned with rabies, such as public-health authorities, veterinary and medical practitioners, livestock organizations, animal protection societies, etc. These groups can provide material assistance to the rabies-control officer by publicizing the programme and otherwise informing the general public whose co-operation must be obtained before specific measures can be successfully applied. If possible, an antirabies campaign should be co-ordinated on a national basis, or at least in adjacent infected areas.

The committee recommends that the following specific measures be applied in affected regions :

- (1) Registration, licensing, and taxation of dogs
- (2) Elimination of stray animals
- (3) Restraint of dogs while the control campaign is under way
- (4) Mass vaccination of dogs free of charge
- (5) Provision of adequate facilities for diagnosis
- (6) Reduction in number of wildlife species where these are a reservoir of the disease
- (7) A continual and energetic publicity campaign.

12. Regional Meetings¹⁰

The committee is in agreement with the almost unanimous opinion expressed in answer to the questionnaire concerning the desirability of regional meetings for the control of rabies. It strongly recommends that WHO arrange regional meetings of appropriate authorities from neighbouring and nearby countries where rabies is a problem so that concerted attacks on this disease will be possible.

It is felt that regional meetings would also provide an opportunity for demonstrating the latest laboratory and field control procedures concerning rabies to technicians wishing to be conversant with recent advances in this field. The committee suggests that WHO make all necessary arrangements for the convening of such meetings when needed.

¹⁰ For resolution adopted by the ad hoc committee of the Executive Board, see section 3, paragraph (3) of footnote 1, page 3.

13. International Conference¹¹

It is the opinion of the committee that an international conference is highly desirable, particularly for the purpose of obtaining general acceptance and wide application of major advances in rabies control.

Promising results have recently been achieved in the laboratory, and field trials of new treatments are under way. The committee recommends that such results be assessed at its proposed second session in 1951, and that at that time a definite recommendation as to the date of such a conference be made.

It is understood that in order to make the necessary arrangements the conference could not take place before 1953.

14. Field Demonstration of Rabies Control in Dogs, Involving the Use of Vaccine

In recent years in several areas of the world, and under specific local conditions, the possibility of rabies control in dogs by the use of vaccine as an adjunct to the usual restrictive measures has been satisfactorily demonstrated. Because of this it is felt that WHO would be making a definite contribution to the control of rabies on an international basis if it were to sponsor a demonstration of a programme of rabies control in dogs in some area where canine rabies is enzootic.

In view of the present availability of a supply of avianized vaccine, the committee recommends the use of this particular type of vaccine in the demonstration. It has been adequately tested for safety in thousands of dogs in the field and shown by thorough laboratory experiments to have high immunizing capacity.

Since this demonstration would be used as a model for application in various parts of the world by WHO, it is essential that the appropriate joint secretary of the committee be given every opportunity to participate in the planning and execution of this demonstration.

The implementation of this recommendation of the committee is left to future exploration on the part of WHO.

15. Collection and Evaluation of Rabies Statistics

The committee wishes to place on record its appreciation of the statistical work carried out under the auspices of the Health Organization of

¹¹ For resolution adopted by the ad hoc committee of the Executive Board, see section 4 of footnote 1, page 3.

the League of Nations, in pursuance of the resolutions of the First International Rabies Conference, held in Paris in 1927.

It considers that the statistical analyses made by McKendrick and Greenwood¹² of the questionnaires provided by rabies institutes throughout the world have consistently indicated the value of the various types of vaccine in use without any significant change from year to year, and therefore that there is no need for continuing with the collection and analysis of the same type of questionnaire.

The committee would, however, be glad if WHO were to prepare for the committee's consideration a simplified form of questionnaire from which indications as to the incidence and social importance of rabies could be obtained.

In this connexion, the committee notes with satisfaction that both the detailed and the intermediate international lists of diseases and causes of death that are to be applied as from 1950 under Regulations No 1 of the World Health Organization provide a separate item for rabies,¹³ so that a much needed improvement of the mortality statistics in respect of rabies is to be expected.

16. Indications for Future Lines of Research¹⁴

Pointers to the more obvious development of research in rabies are interspersed in the subject-matter which constitutes the main observations and recommendations of the committee. However, it is perhaps appropriate to stress several particular investigations on which it would appear to be advisable to direct special attention.

16.1 *Paralysis-producing factor*

The committee is encouraged by the current interest on the part of various groups of workers in the study of the pathogenesis of paralysis produced by sensitization to brain tissue. Isolation of the factor and the determination of its chemical and immunological character might well lead to information applicable to the prevention of the paralytic syndrome occasionally associated with the administration of a course of treatment with rabies vaccines. The development of a technique (see Annex 12)

¹² *Bull. Hlth Org. L.o.N.* 1946, 12, 301

¹³ See World Health Organization (1948) *Manual of the International Statistical Classification of Diseases, Injuries, and Causes of Death*, Geneva, 1, p. 66 (*Bull. World Hlth Org. Suppl. 1*)

¹⁴ For resolution adopted by the ad hoc committee of the Executive Board, see section 3, paragraph (4) of footnote 1, page 3.

for producing a similar syndrome in experimental animals has been the starting-point for research along these lines. Already a method has been evolved in the laboratory for removing this paralysis-producing factor, as exhibited by such animal inoculation, from rabies vaccines, while still preserving their immunizing capacity (see Annex 11). This particular technique needs to be evaluated from the standpoint of its practicability in large-scale production. Further research on the purification of rabies vaccine by various chemical or physical means with the elimination of the paralysis-producing factor is definitely indicated.

Another approach to the elimination of the untoward paralytic symptoms associated with antirabies vaccine treatment, based on the premise that brain-tissue allergy may be a causative mechanism, is the possible substitution of chick-embryo passaged rabies virus as the source of antigen in place of virus-infected brain tissue.

16.2 *Duration of immunity after vaccination with avianized virus*¹⁵

In view of the promising results obtained in the study on the effect of challenge inoculation with street virus in dogs one year after vaccination with avianized virus, the committee deems it advisable to encourage strongly further research on the duration of immunity produced in animals by this type of product. The results of such studies, which can be carried out under laboratory and field conditions, would enable a policy to be formed for the prophylaxis of rabies in dogs.

16.3 *Local treatment of wounds*

The committee suggests that a useful subject of study would be the effects of newer detergents and other agents on the virus of rabies present in the wounds of animals. If on the basis of reliable results any of the substances tested exhibited marked destructive action on the virus, consideration could be given to their possible application to the treatment of wounds inflicted by supposedly rabid animals in human beings.

16.4 *Studies on the characteristics of the virus of rabies*

The committee feels that there is need for the application, to the study of the nature and properties of different strains of the virus of rabies, of some of the more recently developed biophysical and biochemical methods. The important fundamental information provided may well suggest clues to many of the problems at present confronting those responsible for the prevention, control, and eradication of the disease.

¹⁵ For resolution adopted by the ad hoc committee of the Executive Board, see section 3, paragraph (1) of footnote 1, page 3.

16.5 *Ecological studies*

It is now apparent when information is sought on points connected with the epizootiology of rabies that there are many gaps in our knowledge. Reference has been made in the introductory remarks to the extremely interesting observations that have been made on the part played in spreading the disease by such animals as the mongoose and the meercat in the Union of South Africa, and vampire bats in South and Central America, in which areas the disease is enzootic. Reference was also made to the demonstration of the existence of asymptomatic rabies infection in vampire bats. Further surveys are necessary to determine whether the same or other vectors of the disease are not involved. It is found, for example, that rabies appears to recur in particularly well-defined areas in countries in which the disease is enzootic, although the same or similar species of the particular animal demonstrated as a vector of the disease exists in neighbouring territory apparently free of rabies. This may possibly be explained in some measure by the habits of the species concerned, range of movement or migration or, on the other hand, in certain areas there may be special sets of conditions which would explain such facts. Again, there may be other species of animal, e.g., small rodents which are almost unlimited in their variety in certain territories, such as in Africa, which maintain the disease perhaps in an inapparent or unrecognizable form. Skunks, badgers, and ground squirrels have been found to be infected with rabies in South Africa, but at least in the case of the ground squirrel this is due to association in habitat with the yellow mongoose, which is known to have been responsible for 50% of the known cases of rabies in man in the Union of South Africa between the years 1916 and 1947. Associated with these ecological studies should be the pursuance of studies on the characteristics and immunological behaviour, and on apparent species adaptation, of certain strains of the virus of rabies.

Annex 1**NOTES ON METHOD OF PREPARATION OF ULTRAVIOLET
IRRADIATED VACCINE ("UV" VACCINE)***(a) Type of UV apparatus*

Any type using the principle of continuous flow of material with exposure of a thin film should be satisfactory.^{1,4} There are now available two types. One uses a special quartz chamber and special high intensity lamp,³ and the other uses the rotating cylinder method² with an ordinary germicidal type of lamp.^a

(b) Preparation of vaccine^{3,4}

5%, 10%, or 20% suspension of the whole rabies brain may be used, but for each suspension and each apparatus standardization for the time necessary to inactivate virus must be determined. Up to five times this exposure may be given before denaturation of antigen takes place. For routine work, twice the time of inactivating exposure is used.

In the liquid preparation 1 : 10,000 merthiolate is added as a preservative after inactivation. Lyophilization from the frozen state causes an initial drop in potency but then holds well on storage.

References

1. Bozeman, V., Tripp, J. T. & Berry, B. (1950) *J. Immunol.* **64**, 65
2. Habel, K. (1947) *Publ. Hlth Rep., Wash.* **62**, 791
3. Habel, K. & Sockrider, B. T. (1947) *J. Immunol.* **56**, 273
4. Levinson, S. O., Milzer, A., Shaughnessy, H. J., Neal, J. L. & Oppenheimer, F. (1945) *J. Immunol.* **50**, 317

^a Information on the types of apparatus mentioned may be obtained from the Secretary, Expert Committee on Rabies, World Health Organization, Palais des Nations, Geneva, Switzerland.

Annex 2**TECHNIQUE OF PREPARATION OF AVIANIZED
RABIES VACCINE**

Chick embryos, 7 to 9 days old, are inoculated into the yolk-sac with a suspension of Flury^a strain. Ten days later, the embryos are harvested, ground in water, and the suspension centrifuged. The supernatant is

^a Chick-fixed rabies virus passaged subsequently in chick embryos

distributed in phials and lyophilized. The lyophilized preparation is stored at 4°C. At the time of vaccination, the ampoules are rehydrated and the suspension injected intramuscularly in dogs.

Reference

1. Koprowski, H. & Black, J. (1950) *J. Immunol.* **64**, 185

Annex 3

POTENCY MOUSE TEST (HABEL) (FIXED VIRUS)

Sixty Swiss mice, four weeks of age, of either sex or mixed sexes, are vaccinated with 0.25 ml of 0.5% emulsion of vaccine intraperitoneally every second day for 6 doses. Forty similar mice are set aside to serve as controls.

On the 14th day from the first dose of vaccine the mice are challenged with the standard fixed virus strain by intracerebral inoculation. Equal groups of vaccinated mice receive serial dilutions of virus from 10^{-1} through 10^{-5} , and the control group 10^{-4} through 10^{-7} .

The mice are observed for 14 days after receiving the virus. All deaths after the fifth day after inoculation and all mice paralysed at the end of the observation period are counted as rabies deaths. LD_{50} titres are determined in the vaccinated and control groups and the number of ED_{50} determined. The minimum protection required is 1,000 LD_{50} .

Swiss mice need not necessarily be used in the test. Different breeds of mice do vary in their ability to be immunized, but in general any strain of mice of uniform genetic background may be used. The age of the mice is very important, since those under four weeks of age do not immunize well. There is very little difference in mice over the age of four weeks. Therefore it is important that the same strain of mice of the same age be used routinely over a period of time by any one laboratory.

If mice are not available, it is quite possible, to work out a standard procedure along these lines using hamsters as the test animal.

References

1. Habel, K. & Wright, J. T. (1948) *Publ. Hlth Rep., Wash.* **63**, 44
 2. US National Institute of Health (1946) *Minimum requirements: rabies vaccine*, 2nd rev. Bethesda, Md.
-

Annex 4**MODIFICATION OF POTENCY MOUSE TEST (FIXED VIRUS)**

The general principles of the potency mouse test (Habel) as to type and age of animal, number and spacing of vaccine doses, and times and nature of virus challenge all apply to the modified test. The difference consists in the use of groups of animals vaccinated with a series of dilutions of vaccine, all groups being challenged by a single dose of virus.

Groups of 12 mice receive 1.0%, 0.5%, 0.25%, 0.12%, and 0.06% suspension of brain vaccine, and 40 mice are set aside as controls. Fourteen days after the first vaccine dose all vaccinated mice are challenged by the intracerebral inoculation of a virus dilution previously calculated to represent 100 to 1,000 LD₅₀. Control groups of mice are inoculated as in the Habel test, with serial dilution of test virus (10⁻⁴ through 10⁻⁷) in order to determine the LD₅₀ titre.

Using 50% end-point calculations, the actual number of LD₅₀ of virus in the fixed challenge dose is determined, as well as the minimum protective titre of vaccine in immunized mice (dilution of vaccine which protects 50% of the mice). This titre should not exceed the one obtained with 0.5% concentration of vaccine.

Annex 5**STANDARD CHALLENGE VIRUS FOR POTENCY MOUSE TESTS**

Because of demonstrated variations in the properties of strains and substrains of fixed virus, it is necessary that a strain of known invasiveness and antigenic constitution be used uniformly by laboratories in testing the potency of rabies vaccines. Such a strain is now in use in the USA and is available to producing laboratories. In order that this challenge virus should be in approximately the same stage at the time of its use in different laboratories, an arrangement for producing and maintaining virus pools has been established.

A large pool of infected mouse brain is homogenized in a 20% tissue suspension and ampouled in small amounts to be kept frozen at -70°C. This is done in a central distributing laboratory and represents the pool to be used over a period of years. Once a year the central laboratory uses one of these ampoules to produce similarly a secondary smaller pool sufficient to supply all participating laboratories for one year (average of two ampoules per year per laboratory).

This secondary pool is then shipped to each laboratory once a year, in the frozen state, packed in dry ice. On its receipt the laboratory will dilute to a 10^{-3} suspension and inoculate a group of mice intracerebrally, the number of mice depending on the amount of challenge material to be used in the ensuing year. When these mice are down for 24 hours with symptoms of fixed virus rabies, their brains are harvested, homogenized, and diluted to 20% suspension with distilled water. 2.5 ml quanta of the emulsion are placed in each ampoule, which is then flame-sealed and kept frozen until used at -20°C or lower, the lower the better.

Before being used as challenge virus this virus pool should be titrated intracerebrally in six-week-old mice of the same type as those to be used in the potency test — six mice should receive each dilution from 10^{-5} to 10^{-8} . The LD_{50} titre of the virus should fall between dilutions of $10^{-5.5}$ and $10^{-7.5}$. It is usually better to use the contents of two ampoules at each test to obtain more uniform results.

At the end of a year a new sample of seed virus should be obtained from the central laboratory and this procedure repeated.

If conditions under which transportation of the seed virus from the central laboratory to the producing laboratory make it impossible to ship in dry ice, there is no reason why the central laboratory should not maintain its yearly working pool in the lyophilized state.

Annex 6

POTENCY GUINEA-PIG TEST (KOPROWSKI) (STREET VIRUS)

This test was developed for the purpose of evaluation of potency of avianized Flury strain vaccines, because the potency mouse test could not be applied in this case. Guinea-pigs weighing 350-500 g are injected intramuscularly with 0.25 ml of the respective dilution of vaccine (a dilution usually equivalent to a 5% embryo suspension) and three weeks later challenged with street virus together with non-vaccinated control animals. Each animal is injected by the intramuscular route (muscles of one leg) with 0.1 ml of 1:40 dilution (the dilution chosen depends on previous results of titration of the same street virus preparation in guinea-pigs) of canine salivary gland suspension kept as a 20% aqueous suspension at -70°C or lower (temperature of storage is important). The strain of street virus employed is the so called NYC strain, isolated from the salivary gland of a dog dead of rabies in New York City. This strain is maintained by serial passages in dogs injected bilaterally into the masseter

muscles with infected suspension of salivary gland. Material representing fourth to sixth dog passage in the laboratory is employed but there seems to be no contra-indication in using either higher or lower dog passage material. Challenged guinea-pigs are observed for 21 days, and at the present requirement at least 66% of control animals must die of rabies and 80% of vaccinated animals must be protected.

This test may be employed successfully for determination of potency of vaccines containing inactive virus (Semple, ultraviolet irradiated, etc.).

In view of the high susceptibility of hamsters to parenteral infection with street virus, the use of the latter animal may be indicated in cases in which technical difficulties in procuring guinea-pigs may be encountered.

Annex 7

PREPARATION OF HYPERIMMUNE ANTIRABIES SERUM

Either rabbits or sheep were immunized by means of repeated injection of fixed strains of rabies. The animals were bled at regular time intervals and large pools of serum or plasma prepared. These were fractionated (by such substances as ammonium sulfate, sodium sulfate, or methanol), and the gamma, or beta and gamma, globulin fractions concentrated.

Hyperimmune serum concentrates have been tested in neutralization tests in mice in which several dilutions of antiserum concentrates were mixed with one dilution of fixed virus containing approximately 100 LD₅₀. These mixtures were inoculated intracerebrally in mice and the minimum protective titres (final) of concentrates calculated.

In the protection test in hamsters, serum concentrates were administered after inoculation of the animals with street virus.

Both the mouse neutralization and the hamster protection test indicated a high content of neutralizing antibodies. In repeated tests the serum neutralizing antibody titre has been shown to be over 1 : 100.

References

1. Habel, K. (1945) *Publ. Hlth Rep., Wash.* **60**, 545
 2. Koprowski, H., Scheer, J. van der & Black, J. (1950) *Amer. J. Med.* **8**, 412
-

Annex 8

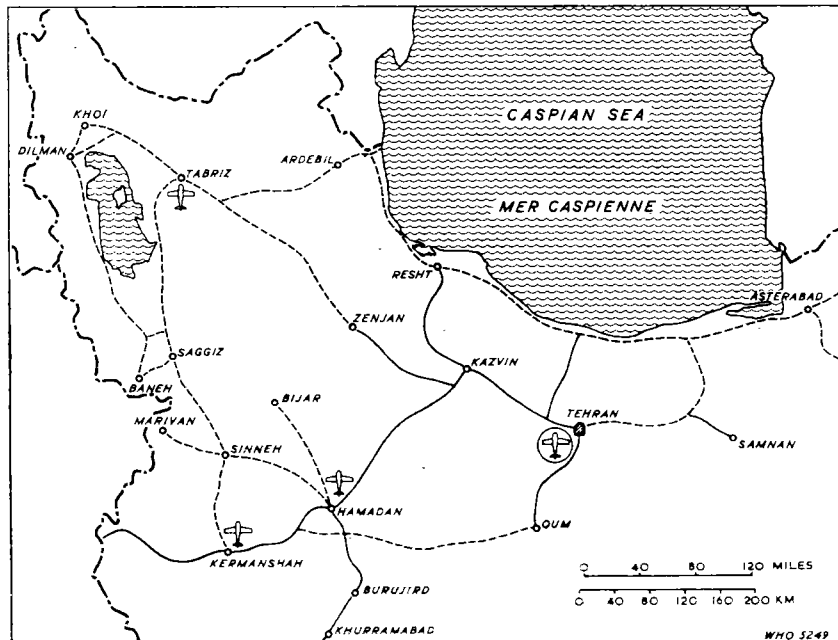
FEASIBILITY OF A TRIAL OF HYPERIMMUNE SERUM IN MAN
Cases of Rabid-wolf Bites in Iran

The frequency of rabid-wolf bites in man is relatively high in Iran — 60 in 1949, 22 in the period from January to April 1950 — and these are always extremely severe.



Cases always occur in groups: in 1949, 12, 19, 26, and 3; in 1950, 14 and 8. At least half of the number of persons in each group exhibit mutilating head and face wounds. The case fatality rate exceeds 30% in spite of intensive treatment by a Fermi type vaccine.

Wolves still roam widely in Iran. The sketch map which accompanies this Annex (fig. 1) indicates the roads by which it will be possible to reach human cases of rabies due to wolf bite at the latest in 48 hours. The three aerodromes are also indicated on the map.

FIG. 1. MAIN LINES OF COMMUNICATION IN NORTH-WESTERN IRAN



— hard surface roads
 ---- other main roads

 main airport
 secondary airport

Arrangements will be made, by approach to the Minister of the Interior, for precise instructions to be issued to all officials concerned in the area, so that persons bitten by wolves will be transported to the nearest negotiable road. At the same time a telegram will be specially expressed to the Institut Pasteur to advise on the point at which the patient can be picked up.

Annex 9

**FIELD TRIAL OF ANTIRABIES HYPERIMMUNE SERUM
IN HUMAN BEINGS EXPOSED TO RABID-WOLF BITES**

The committee decided the following conditions for this trial should apply :

- (1) Wolf bites within a series to be treated on a strictly alternating basis with
 - (a) hyperimmune serum and vaccine, and
 - (b) vaccine alone.
- (2) The first treatment in either series not to be given later than 72 hours after the bite.
- (3) The dosage of serum to be 1 ml per kg body-weight injected intramuscularly in the buttock.
- (4) The vaccine to be the one currently employed at the Institut, i.e., 5% sheep-brain phenol attenuated vaccine (Fermi).
- (5) The dosage of vaccine to be 21 injections of 5 ml, given daily.
- (6) The first injection of vaccine
 - (a) in the case of serum plus vaccine treatment, to be given 48 hours after the serum injection, and
 - (b) in the cases treated with vaccine alone, to be given at the same time as serum is administered to alternate cases.
- (7) A 20 ml sample of each vaccine batch to be used for treatment of cases in the field trial to be sent by air mail to Dr Karl Habel, Chief, Laboratory of Infectious Diseases, Microbiological Institute, National Institutes of Health (US Public Health Service), Bethesda, Md., USA, for potency testing. In addition, a sample of fixed virus production strain to be supplied.
- (8) Only persons severely bitten by wolves to be included in this trial.

(9) Local treatment in the form of thorough cleansing with soap solution to be applied in every case. In no instance is serum infiltration of the bite wound to be undertaken.

(10) In every instance, all points of the case-history sheet to be filled in.

(11) The patients to be kept under direct supervision for as long as possible, but at least for 30 days after termination of treatment.

(12) The patients to be kept under general observation for a period of six months after the bite. A male nurse to be stationed at the place where the patients are living.

(13) Samples of blood for serum to be collected from patients of both series not earlier than 24 hours after antirabies serum injection, but before the first dose of vaccine, again on the 15th day, and finally on the 30th day after initiation of vaccine treatment.

(14) 5 ml of each serum sample drawn from the patients to be sent by air mail to Dr Karl Habel for neutralization tests.

(15) Diagnostic procedures to include pathological examination and attempts at recovery of virus.

Annex 10

SUGGESTED TECHNIQUE OF BIOLOGICAL TEST FOR CONFIRMATORY DIAGNOSIS OF RABIES

At least 6 mice should be inoculated intracerebrally with a 10% suspension of the tissue to be tested. The animals should be observed for at least 30 days and any animal developing symptoms of infection should be killed and the brain examined for Negri bodies.

The use of antibiotics to diminish risks with bacterial contaminants is recommended when necessary (penicillin 500 to 1,000 units per ml plus streptomycin 50 to 100 units per ml).

When mice are not readily procurable at least three hamsters may be substituted for inoculation intracerebrally.

Reference

1. Johnson, H. N. & Selters, T. F. (1948) *Rabies*. In: American Public Health Association, *Diagnostic procedures for virus and rickettsial diseases*, New York, p. 219
-

Annex 11**PREPARATION OF RABIES VACCINE WITH REMOVAL OF PARALYSIS-PRODUCING FACTOR**

Shell-frozen 20% whole suspension is lyophilized. Benzene is added, the dried material is emulsified, and the virus inactivated by the benzene for 12 hours at 56° C. The benzene is then decanted and the residue removed by ether treatment. The dry sediment is suspended in 0.1 M calcium acetate and allowed to stand for several hours in the cold. Filtration or centrifugation then separates the sediment, which is saved, and the supernatant is discarded. Several washings of the sediment with distilled water are made before the sediment is finally suspended in water.

Reference

1. Bell, J. F., Wright, J. T. & Habel, K. (1949) *Proc. Soc. exp. Biol., N.Y.* **70**, 457

Annex 12**GUINEA-PIG TEST FOR PARALYSIS-PRODUCING FACTOR**

A mixture of the following ingredients is made by forcibly filling and emptying a syringe :

- (a) One part of 20% brain suspension
- (b) One part of "Falba",^a lanolin, or "Arlacel"^a
- (c) Two parts of mineral oil containing 1 mg of killed BCG or *Mycobacterium butyricum*

1 ml of this mixture is inoculated into the muscles of the shoulder of guinea-pigs weighing 200 to 250 g. Animals are observed for paralysis for 60 days, usually develop symptoms by the 30th day, and all survivors are checked by histopathological examination.

References

1. Freund, J., Stern, E. R. & Pisani, T. M. (1947) *J. Immunol.* **57**, 179
2. Freund, J., Thomson, K. J., Hough, H. B., Sommer, H. E. & Pisani, T. M. (1948) *J. Immunol.* **60**, 383

^a Proprietary products

**WORLD HEALTH ORGANIZATION
TECHNICAL REPORT SERIES**

	Number		Price	
Antibiotics, Expert Committee on				
Report on the first session	26	9d.	\$0.10	Sw. fr. 0.40
Bilharziasis, Expert Committee on				
First report	65	2/3	\$0.30	Sw. fr. 1.20
Bilharziasis in Africa, Joint OIHP/WHO Study-Group on				
Report on the first session	17	9d.	\$0.10	Sw. fr. 0.40
Biological Standardization, Expert Committee on				
Report on the third session	2	1/6	\$0.20	Sw. fr. 0.80
Report on the fourth session	36	9d.	\$0.10	Sw. fr. 0.40
Fifth report	56	1/3	\$0.15	Sw. fr. 0.60
Sixth report	68	1/6	\$0.20	Sw. fr. 0.80
Report of the Subcommittee on Fat-Soluble Vitamins	3	9d.	\$0.10	Sw. fr. 0.40
Brucellosis, Joint FAO/WHO Expert Committee on				
Report on the first session	37	2/-	\$0.25	Sw. fr. 1.—
Second report	67	2/-	\$0.25	Sw. fr. 1.—
Cholera, Expert Committee on				
First report	52	1/3	\$0.15	Sw. fr. 0.60
Cholera, Joint OIHP/WHO Study-Group on				
Report on the third session	18	1/3	\$0.15	Sw. fr. 0.60
Communicable Diseases of Childhood, Active immunization against common				
Report of a group of consultants	6	1/3	\$0.15	Sw. fr. 0.60
Diphtheria and Pertussis Vaccination				
Report of a conference of heads of laboratories producing diphtheria and pertussis vaccines	61	4/3	\$0.55	Sw. fr. 2.20
Drugs Liable to Produce Addiction, Expert Committee on				
Report on the second session	21	9d.	\$0.10	Sw. fr. 0.40
Third report	57	9d.	\$0.10	Sw. fr. 0.40
Environmental Sanitation, Expert Committee on				
Report on the first session	10	2/-	\$0.25	Sw. fr. 1.—
Second report	47	1/3	\$0.15	Sw. fr. 0.60
Health Statistics, Expert Committee on				
Report on the first session	5	9d.	\$0.10	Sw. fr. 0.40
Report on the second session (including reports on the first sessions of the Subcommittees on the Definition of Stillbirth and Abortion, on the Registration of Cases of Cancer as well as their Statistical Presentation, and on Hospital Statistics)	25	2/-	\$0.25	Sw. fr. 1.—
Third report (including second report of the Subcommittee on the Registration of Cases of Cancer as well as their Statistical Presentation)	53	2/9	\$0.35	Sw. fr. 1.40

	Number		Price	
Hepatitis, Expert Committee on				
First report	62	1/6	\$0.20	Sw. fr. 0.80
Hygiene of Seafarers, Joint ILO/WHO Committee on				
Report on the first session	20	9d.	\$0.10	Sw. fr. 0.40
Influenza, Expert Committee on				
First report	64	1/6	\$0.20	Sw. fr. 0.80
Insecticides, Expert Committee on				
Report on the first session	4	2/3	\$0.30	Sw. fr. 1.20
Report on the second session	34	4/3	\$0.55	Sw. fr. 2.20
Third report	46	2/-	\$0.25	Sw. fr. 1.—
Fourth report	54	5/-	\$0.65	Sw. fr. 2.60
International Pharmacopoeia, Expert Committee on the (formerly Expert Committee on the Unification of Pharmacopoeias)				
Report on the fourth session	1	9d.	\$0.10	Sw. fr. 0.40
Report on the fifth session	12	9d.	\$0.10	Sw. fr. 0.40
Report on the sixth session	29	1/3	\$0.15	Sw. fr. 0.60
Report on the seventh session (including report on the first session of the Subcommittee on Non-Proprietary Names)	35	1/6	\$0.20	Sw. fr. 0.80
Report on the eighth session (including report on the second session of the Subcommittee on Non-Proprietary Names)	43	2/-	\$0.25	Sw. fr. 1.—
Ninth report (including third report of the Subcommittee on Non-Proprietary Names)	50	2/-	\$0.25	Sw. fr. 1.—
International Sanitary Regulations				
World Health Organization Regulations No. 2	41	5/-	\$0.65	Sw. fr. 2.60
Malaria, Expert Committee on				
Report on the third session	8	2/3	\$0.30	Sw. fr. 1.20
Report on the fourth session	39	1/6	\$0.20	Sw. fr. 0.80
Malaria Conference in Equatorial Africa				
Report	38	3/6	\$0.45	Sw. fr. 1.80
Malnutrition, Prevention and treatment of severe, in times of disaster				
Report of a group of consultants	45	2/9	\$0.35	Sw. fr. 1.40
Maternity Care, Expert Committee on				
First report: a preliminary survey	51	1/3	\$0.15	Sw. fr. 0.60
Mental Health, Expert Committee on				
Report on the first session	9	2/3	\$0.30	Sw. fr. 1.20
Report on the second session	31	2/9	\$0.35	Sw. fr. 1.40
Alcoholism Subcommittee				
Report on the first session	42	1/3	\$0.15	Sw. fr. 0.60
Second report	48	2/-	\$0.25	Sw. fr. 1.—
Nursing, Expert Committee on				
Report on the first session	24	1/6	\$0.20	Sw. fr. 0.80
Second report	49	1/3	\$0.15	Sw. fr. 0.60
Nursing Education, Working Conference on				
Report	60	1/6	\$0.20	Sw. fr. 0.80