DR. F. M. ALLEN, New York: I do not believe anything can be said offhand on a subject as little understood as the blood lipoids. A subject is always opened up when the necessary methods become available, and it is to be hoped that the methods of Bloor will serve to bring light on these problems. These extensive observations of Drs. Joslin and Gray are important pioneer work on the clinical side. A fundamental question back of these phenomena is why fat behaves in this way in diabetes. Evidently it is one more illustration of the fact that diabetes is an impaired assimilation of all foods, and not merely of carbohydrate. But the mechanism by which fat is normally taken up from the blood, and which breaks down in diabetes, is unknown. We have some animal experiments in progress which may later yield a little information concerning it. One point which may perhaps be shown when Dr. Joslin's full figures are published is the possible influence of lipemia on the carbon dioxide capacity of the blood. When the plasma is so full of fat, what is the relation between its carbon dioxide capacity and the values in the alveolar air? Therapeutically, it is always desirable, and generally possible, to clear up a pathologic lipemia. We have had some patients in whom heavy lipemia was thus cleared up and who are alive now. High lipemia was formerly regarded as a very bad prognostic sign, and presumably an intractable lipemia may still be so considered. Dr. Geyelin has had some of the severest cases of diabetes ever described. The one which he reported with Dr. Du Bois was perhaps the severest ever studied by accurate methods. There is a possible question whether the metabolism is in some cases specially defective on the fat side and in other cases on the carbohydrate side. Distinctions in types of diabetes have generally proved unreal. Probably the apparent differences in lipemia and other symptoms will be explainable by diet and other special influences.

DR. HORACE GRAY, Boston: Dr. Geyelin's point, as I understand it, is that two of his patients, as they improved, lost their cloudy plasma, while another patient, who improved little, kept the cloudiness. That can perhaps be reconciled with our figures by saying that although, in general, it is distinctly not safe to judge the amount of fatty substances in the blood by the appearance of the plasma, still the very creamy plasmas do have high lipoids. Stating this in other words, we have described plasmas in twelve divisions, from "clear" to "thick creamy." In the lower divisions up through "very cloudy" it is a delusion to think one can predict whether the lipoids will be high or low, but the plasmas which were described as "milky," "creamy," and "thick creamy" did regularly have a high lipid content.

Dr. Allen's point about bad prognosis in patients with creamy plasmas agrees with Dr. Joslin's patient, Case 786, who had thick creamy plasma and total lipoids of about 16 per cent. It may interest you to know incidentally that the highest value generally accepted in the German literature is about 19 per cent., while there have been reported even higher.

When you start to do this work two things need to be agreed on: the method and the nomenclature. The naming is a complicated business, but one thing we can easily do is to use "lipemia" in a more definite way. To some people it means a cloudy plasma; to others it means a large lipid percentage on analysis. We might well adopt the French "lactescence" for the gross appearance, and restrict "lipemia" or "hyperlipemia" to the chemical analysis, similar to the use of the term "hyperglycemia." The method which Dr. Joslin has recommended under the name of "Bloor's fat method" was called by Dr. Bloor "total-fat." Dr. Joslin and I venture to urge this change of name to avoid confusion with "total lipoids," which, being a more inclusive term, seems more entitled to be called "total."

For a routine practice the use of whole blood instead of plasma is easier and requires less blood.

Social Reform.—All attempts at reform at present should be in the direction of acquiring greater flexibility in the social organism, not in the direction of a definite system.—*Scientific American.*

STANDARDIZATION OF SERUMS AND VACCINES

SOME DIFFICULTIES AND MISUNDERSTANDINGS

G. W. McCOY

Director, Hygienic Laboratory, U. S. P. H. S.

WASHINGTON, D. C.

The misunderstanding that exists even among usually well informed health officers and physicians in regard to the standardization of biologic products is sufficient reason for a brief consideration of the subject. For example, under the heading "Standardization of Biological Products" on page 1416 of THE JOURNAL, May 12, 1917, a note states that the official Conference of State and Territorial Health Officers adopted a resolution recommending that the Public Health Service "establish and promulgate standards as to potency with reference to all biologic products, vaccines and serums, not only with relation to diphtheria and tetanus antitoxin, but especially with relation to antimeningococcus and pneumonia serums, and vaccines for typhoid fever, paratyphoid fever, and bacillary dysentery."

A standard is defined as "that which is set up and established by authority as a rule for the measure of quantity, weight, extent, value or quality; especially the original specimen weight or measure sanctioned by government as the standard pound, gallon, yard, width, or the like."

One sees that there are strict limitations as to what may be allowable under the definition of a standard. Those who have devoted the most time and have had imposed on them the greatest responsibilities in relation to the field of biologic products have thus far been willing to recognize as standardizable only a small number of preparations. Firmly based on substantial scientific grounds, standards have been formulated for the antitoxins of diphtheria and tetanus. In the case of the former we have as a standard a serum which laboratory tests and long observation have shown to be constant, a true standard by which specimens of antidiphtheria serum can be measured, though perhaps not quite so accurately as we can measure things which do not depend on living creatures (guinea-pigs) for the indications that the end of the measure has been reached. With slight modifications, what has been said of diphtheria antitoxin may be said of tetanus antitoxin. It is not the purpose to go into the details of the beautifully worked out methods of testing these antitoxins, but the standards have now been in use for a number of years, and are very satisfactory; though unless large numbers of test animals are used, only approximate measurements of the strength (unitage) of given specimens of serum may be made. A most important point is the fact that the measure of the antitoxic value of these serums is also a measure of the usefulness of the product in clinical use.

At the Hygienic Laboratory, much time has been devoted also to the standardization of antityphoid vaccine, and there has been devised a tentative standard for this agent. This tentative standard requires that the dose intended for human beings, when administered to rabbits in a certain manner, shall provoke a certain agglutinin response. It was recognized that the agglutinin response was not necessarily a measure of the prophylactic value of the vaccine as applied to man, but it was felt that the procedure would improve the quality of the vaccine on the market, which had

been found to be very variable. Experience has shown that it is advisable to furnish a standard vaccine and to require that commercial products shall equal that in agglutinin producing properties; in this manner, inequalities in results, due to differences in technic, are eliminated.

This completes the list of preparations for which there are standards, only a small fraction of the great number of biologic products on the market. This does not mean that attempts at standardizing other products have not been made. It simply means that science has not progressed far enough to enable the proper authorities to set up a rule for the measurement of preparations which are of recognized value.

There are three fundamental difficulties in standardizing biologic products. The first of these is the fact that none of the factors is constant. We are dealing with living organisms in which vital processes are going on, or with the products of these vital processes, and we know that neither processes nor products are constant. It is true that certain of the toxins and antitoxins are relatively stable, when kept under certain conditions, and this stability was at once utilized to establish such standards as we now have.

The second great difficulty is the fact that animals must be used in making comparisons between the standard and the preparation to be investigated. Individual laboratory animals vary in their reactions to toxic agents to which they are susceptible, as well as to the antitoxic agents, these terms being used in a broad sense.

The main difficulty, however, is the fact that we lack laboratory animals susceptible to many of the agents which we need to test; thus, the ordinary staphylococcus, the gonococcus, and the pertussis bacillus are good examples of organisms which we cannot submit to animal tests.

These points make it clear that, in the very nature of things, we cannot hope to have standards which are comparable to those of a purely physical nature, or even to those having a chemical basis.

When a preparation is in the purely experimental stage, that is, before clear evidence as to its value is available, it would be a waste of time and energy to try to standardize it officially; and the same may be said of products for the value of which there is no substantial basis.

Certain preparations which have passed the experimental stage within recent years, and certain others the value of which has long been established, are without recognized standards, as will be shown presently.

Aside from the purely technical difficulty of standardizing many preparations is the fact that the establishment of an official standard is likely to be regarded as setting the stamp of governmental approval on claims made for the usefulness of the preparations. It would certainly be contrary to public interest if official action capable of such interpretation should be taken. The German authorities have established a number of provisional standards for biologic preparations, but the wisdom of the policy is debatable.

SPECIAL PREPARATIONS

Some of the special preparations that have been mentioned as being in need of standardization, a need that no one will deny, will be considered.

Antimeningococcus Serum.—The effectiveness of this agent seems to be established, but experience has shown that at least some specimens of serum have

been found to be useless in certain outbreaks of meningitis. One reason for this, of course, is that, from an immunologic point of view, there is a very considerable variation in the organisms isolated from various sources. The meningococci have been divided into groups depending on their serologic reactions, but there is no agreement among workers as to the number of groups nor the characteristics which govern the differentiation of the several groups. All antimeningococcic serums are polyvalent, that is, contain antibodies for various cultures of the organism; but just what constitutes sufficient polyvalency is not determined. It would not be sufficient to say that a serum shall represent a minimum number of different cultures, because this might overlook whole groups; nor would it suffice to say that all groups of the organism must be represented, although this is inquired into during governmental inspections, because of the lack of agreement as to grouping and the fact that immunologic relations of individual members of groups vary considerably. These difficulties have not deterred those who are interested in the subject from endeavoring to establish some criterion of the value of the serum.

All serums made in the United States are submitted to some form or other of potency test. Several manufacturers determine the agglutination titer of the serum, others, the titer in a complement deviation complex, while others make an opsonic index determination. An animal protection test is also in use. Unfortunately, there is no agreement among those who have given this subject special attention as to which test is the more satisfactory; no proposed standard has sufficiently impressed those who are concerned with this problem to lead to its adoption.

Good authorities insist that the results of the therapeutic use of the serum afford the only reliable evidence of its value, and it may be remarked that almost any manufacturer can produce glowing testimonials as to the clinical results of the use of his product. The question that naturally occurs is why some one of these tests is not adopted, and all serums required to comply with it. The answer is that in spite of much experimental work we are without sufficient evidence as to the superiority of any method of testing. The multiplicity of types of meningococci, and the variations even among members of the same group of organisms, make accurate standardization impossible at the present time.

The serums made by American manufacturers represent all the way from ten to forty-six strains of meningococci, and most manufacturers make it a rule to utilize every new strain that comes into their possession unless it is one that is immunologically identical with cultures they are using already.

Finally, an additional obstacle to the standardization of the product is the fact that, even when all conditions are favorable, it is difficult for two workers, using the same method, to get comparable results.

Antipneumococcus Serum.—It is only recently that the value of serum in this disease has been put on a sufficiently substantial foundation to justify its general use. Indeed, this serum illustrates the creation of a demand prior to the placing of the product on a sound scientific basis. As soon as it became known that there was in experimental use a promising treatment for pneumonia, physicians naturally desired to utilize it in a disease otherwise most unsatisfactory from a therapeutic point of view; and manufacturers sought

to meet the demand. In the case of pneumonia we have conditions somewhat analogous to those in meningococcus infections, though probably less complex. The causative organisms of pneumonia can be divided immunologically into several groups; four, according to Cole and his collaborators.

At the present time there is evidence of the therapeutic value of a serum prepared against but one of the types (Group I). While scientific observation has failed to show the value of specific serum against the other groups (II, III, IV), or of Group I serum against these other groups, clinicians working without group determinations and presumably without adequately controlling their work, have reported satisfactory results from the use of the serum in pneumonia in general. We may be pardoned if we remain skeptical as to the validity of the observations.

Antipneumococcus serum affords a good example of the difficulties in standardization when what we might call the "test object" is a living organism. It is impossible to secure a constant minimum lethal dose of culture as a criterion. The rate of growth varies, and the virulence varies. With these factors so inconstant, and yet absolutely vital, there seems to be no immediate prospect beyond the establishment of a provisional standard.

Other Serums.—When we come to consider these, we find that there is practically no ground for establishing standards. To say that they should show some evidence of potency, without prescribing a minimum, is futile; to prescribe a minimum that would not be reasonably near the maximum obtainable would put a premium on mediocre work on the part of producers; and we have no means of demonstrating the maximum degree of potency.

Bacterial Vaccines.—It is perhaps not too sweeping to say that at present, as regards the majority of bacterial vaccines, potency standardization is both impracticable and undesirable. Aside from antityphoid vaccine, evidence of value is meager, and practically none of the vaccines can be submitted to potency tests on laboratory animals. Generally speaking, the doses that have been adopted are purely arbitrary. In many cases the dose might be multiplied by ten, or divided by the same factor, without, so far as we know, influencing the effect of the vaccine.

The determination of the number of organisms per given volume of fluid is apparently a simple procedure; but, as a matter of fact, two workers equally careful will often get most divergent results; even when the same method is employed, and when different methods are used, the results are little better than guesses based on turbidity would be. Obviously this is a minor matter, in view of what has been said above regarding the size of dose.

The question of standardization of tuberculins is one that recurs constantly. These agents are employed comparatively little in the treatment of tuberculous infections, but they are used rather extensively in diagnosis. The varieties are legion. It would scarcely serve merely to require that they shall produce a reaction, unless we prescribe the quantity and the dilution. Nor do we know whether or not the results of animal experiments are comparable with the results that may be expected when the agent is applied to man.

Vaccine Virus.—The ultimate criterion of the potency of this agent is the percentage of "takes" it will give, when used to vaccinate persons not pre-

viously successfully vaccinated. Whether any testing on laboratory animals can be used as a satisfactory substitute for this is uncertain.

Even here there must be qualifications, if we are to speak in terms of a standard; thus, how large an abrasion is required to give a successful result, and how much may the vaccine be diluted and still give uniform success? This particular product does not especially concern those of us who are familiar with the subject of standardization; because the demand for vaccine virus comes largely from health officials who are in position to judge from the results of use as to the potency of a given manufacturer's product. If the vaccinator gets poor results he is not likely to order a second supply from the same source. In other words, a poor product will not be a commercial success. It is the judgment of those who have had most experience with the subject that we should not at present insist on a vaccine which is free from contaminating micro-organisms, though the promising work of Noguchi leads us to hope that we may have a dependable pure vaccine in the not far distant future.

Antirabic Virus.—So long as the preparation of this agent followed, or followed approximately, the original Pasteur method, the products probably varied comparatively little. Now, however, there are available dialyzed and desiccated viruses for which certain advantages are claimed; attempts at standardizing these have been made, but there is not sufficient evidence to warrant the adoption of a standard.

In connection with vaccine virus and antirabic virus, there is this element of security in regard to potency: The propagation of the product requires the inoculation of material from animal to animal; and the failure in one case to develop pocks, and in the other the failure to produce rabies, would indicate at once that the material has become inert; in either case, failure to infect animals used for propagation may be regarded as evidence of the lack of power to protect man.

Many products may be tested by purely serologic methods, but the results of such tests may or may not bear any relation to the usefulness of the product. Of what value is it to know that a certain serum gives a reaction in a certain dilution by means of complement deviation, agglutination, opsonic index determination, or a bacteriolytic determination, when we do not know that these reactions bear any relation to one another; or, what is more important, to the effect the serum may have on the infection against which it is expected to act?

A possibility that must not be lost sight of is that the establishment of a standard may result in the fixing in the minds of workers and manufacturers the feeling that, as regards the particular product, there is no need for improvement. If a thing conforms to a government standard, why try to improve it?

A factor that must be considered in discussing the standardization of biologic products is the rate of deterioration. It is not too much to say that we know almost nothing of this, barring the two antitoxins for which we have potency standards. This absence of information has led to great variation in "expiration" dating for the same product. To remedy this, some manufacturers place the date of manufacture on the label, leaving to the purchaser the determination of the wisdom of using the preparation. There is justification for this, on account of the fact that the man-

ner in which products are kept influences deterioration. The serum or vaccine displayed on the pharmacist's shelves, or stored in a drawer, loses its efficacy much faster than one kept in an adequate refrigerating device. If the purchaser knows the age of a preparation, he may be in some position to judge of its probable value, when kept under given conditions.

SUMMARY

From the foregoing it is evident that these questions have already claimed much attention, and that the subject of standardization may be thus summarized:

The chief difficulty in standardizing biologic products is due to the unstable nature of the factors.

Legal standards have been established for diphtheria antitoxin and tetanus antitoxin. A provisional standard has been established for antityphoid vaccine.

Vaccine virus and antirabic virus are, in effect, tested for potency in the process of manufacture.

It is not wise to attempt to standardize products in the purely experimental stage, or those for which there is no sound scientific basis.

Those who are engaged in manufacturing and controlling these products are alive to the necessity for standardization.

COMPARISON OF METHODS OF
TREATING WOUNDS

KENNETH TAYLOR, M.A., M.D.

Director, the Robert Walton Goelet Research Fund. American Red
Cross Hospital of Paris

PARIS, FRANCE

The difficulty of judging and comparing the clinical results of different methods of the treatment of wounds is so great that it seems desirable to consider how more exact information as to the value of a treatment can be attained. It is obvious from a study of the literature since the beginning of the war that the superiority of no one form of treatment has become firmly established. The treatment of infection in wounds is thus still largely in the experimental stage.

The haphazard way of trying first one and then another method on each case, hoping to stumble on a suitable one, is as nearly barren of progress as is treating all wounds in the same way. This proceeding is not likely to lead to the recognition of the best methods of treatment for infected wounds. It is important, therefore, to organize in hospitals a system which may give a clear idea of what is being accomplished by each method of treatment.

As in most other therapeutic problems, the great difficulty in associating cause and effect lies in the number of variable factors which must be controlled. In the case of infected wounds, the first and one of the most important of these factors is the personal element in the manner of applying the treatment and handling the wound. It is of little use to compare two methods of treatment conducted by different men. No two surgeons will carry out the details of the same treatment in exactly the same way. One may secure satisfactory results by a certain method entirely from delicate and careful handling of the regenerating tissues of the wound, while another, using the same method of dressing, may counteract the benefits of the

system by careless treatment of the young epithelium and granulations with which he is dealing. Yet both results may be ascribed to the particular method or dressing solution which is being used.

It is also unsatisfactory to compare the results of one method of treatment conducted in a series of cases with a different method used by the same surgeon in a previous series. It will often be stated that better results were obtained with the second method than with the first, when the difference is really due, not to a better method, but to the increased experience of wounds that has been gained by the surgeon.

There are other important factors which vary with the patient. First there is that of the *general vitality* and ability to react to the infection. Although this is not so great as might be supposed, owing to the fact that much of the military surgery deals with men more or less carefully selected for their sound health and freedom from debilitating disease, it cannot be entirely discounted.

The *age of the wound* and the previous treatment to which it has been subjected are also factors which must be standardized to be compared. This can partly be accomplished by establishing a system of periodic examinations and descriptions of each case at short intervals, and grouping the cases according to the number of *days since the injury* was received.

One of the most serious difficulties is encountered in the varying conditions within the wounds themselves. No two wounds can be safely considered to be exactly alike, although their external appearance may be very similar. Differences of *location and vascularity* may influence seriously the course of repair. The degree of *trauma*, both the obvious damage and the more subtle microscopic destruction of the soft tissues, is probably the most important of all the factors to standardize and yet the most difficult to estimate. Thus compound fractures are particularly unsatisfactory for study. The presence of *foreign bodies*, especially fragments of clothing and dirt, loose pieces of bone, etc., also alters materially the rate of repair; and, finally, the degree and kind of *infection*, which may vary with each patient, still further complicates the problem.

Although the difficulties of comparison are, as may be seen, great, there are certain methods by which cases can be arranged to reduce the number of the variable factors which confuse the issue and to give a more accurate comparison between them.

There are three ways in which these cases can be profitably studied. The first is by comparison of groups of selected cases. Each surgeon may collect under his service cases which have some points in common—for instance, open fractures, wounds of soft parts only, or cases of approximately the same age as to the injury. These may then be divided into groups, each set being treated in a different way.

Another system lies in a comparison of large groups of unselected cases. This is a logical but cumbersome method. The beds of the service may be divided into two or more groups, each treated by a different method, and patients sent to each group on entry, in rotation. Thus there would be no choice of treatment for a particular case as the result of a preconceived idea of suitability, but the distribution would be purely automatic. Owing to the great variety of patients on each method of treatment, it is necessary to continue the system for a long period of time and in a large number of cases in order to form any satisfac-