Other methods employed in the early part of the nineteenth century were the use of bits of thread or silk saturated with vaccine lymph, and these were drawn under the skin and allowed to remain till infection took place.

Bits of goose quill were also sharpened like a pen and then charged with lymph. These were often used for vaccinating by puncture.

(*c*) *Dried Vaccine Lymph in the Form of Powder.*—This method of using vaccine lymph has been employed to some extent, but has not come into general use. M. Ciaudo says he has obtained good results with dried vesicles after from one hundred and ten to one hundred and thirty days. Warlomont experimented with vesicles cut into small fragments and rapidly dried in a drying apparatus. These fragments at the end of two or three days were reduced to a fine powder and preserved in tubes closed only with cotton. When the powder was used in this condition the vaccinations were not very successful; but if the powder was allowed to macerate in glycerin and water for twenty-four hours (thus allowing the vaccine microbes to pass from a latent state to one of active life) and was then used, good results were obtained, even though the powder was more than three months old.

2. *Vaccine Lymph in the Moist Condition—Pulp.*—This method, first introduced by Ciaudo, of Milan, consists in excising the vesicles, freeing them from all detritus, making a homogeneous paste, and adding to each vesicle 0.5 gm. of chemically pure glycerin; this is placed in a small glass vial, and covered with more glycerin to exclude the air.

Vaccine material thus prepared preserves its activity for a long period, and is very efficient. Its effects are, however, occasionally quite irritant. A similar mode is adopted in Holland, the pulp being preserved between glass plates.

In Germany it is customary to employ capillary tubes for single vaccinations, but when large quantities are required, the glycerinated emulsion is put into small vials holding enough for twenty-five to fifty or one hundred vaccinations in each. Glass plates, slightly excavated and fitted with tight covers, are also in use.

Capillary Tubes.—These were first brought to public notice by Dr. Husband, of Edinburgh. Their essential characteristics were stated to be as follows: (1) They should be so slender as to admit of ready sealing in the flame of a lamp; (2) they should be large enough to contain lymph sufficient at least for one vaccination; (3) they should be long enough to admit of sealing without subjecting their contents to the heat of the flame; (4) they should be strong enough to admit of handling without breakage. Dr. Husband advised the following dimensions: Average length, 2¾–3 in. (about 7–8 cm.); diameter, $\frac{1}{25}$ in. (about 1 mm.). Another form is made with an enlargement at the centre of the tube. Tubes are sometimes made with closed ends, that the interior may be kept clean until ready for use. When required for use the ends should be broken off, a small rubber bulb applied, and the lymph should be blown out upon a glass plate, from which it may be conveyed with the lancet to the scarifications. Tubes should be used only for the preservation or storage of glycerinated lymph.

Glycerinated Lymph.—As early as 1850 Mr. R. Cheyne[14] had called attention to the superiority of vaccine lymph which has been treated with glycerin. Curschman, however, credits Müller of Berlin with this discovery. Warlomont of Brussels refers to the subject in his "Traité de la Vaccine" and had patented a process of such mixture in 1882.

The German Government also recognizes this process in the regulations issued April 28th, 1887.

Chambon, Menard and Straus,[15] of Paris, also by means of experiment, arrived at similar conclusions. So also did Leoni, Klein, Kitasato, and others.

Dr. Copeman[12] sums up the advantages of glycerinated lymph as follows:

1. By employing the method of glycerination of lymph pulp great increase in quantity can be obtained without any consequent deterioration in quality, the percentage of insertion success following in its use being equal to that obtained with perfectly active fresh lymph.

2. Glycerinated lymph does not dry up rapidly, as does unglycerinated lymph, thus simplifying the process of vaccination.

3. Glycerinated lymph does not coagulate, so that it never becomes necessary to discard a tube on this account.

4. Glycerinated lymph can be produced absolutely free from the various streptococci and staphylococci which are usually to be found in untreated calf lymph, and which are, under certain circumstances, liable to occasion suppuration.

5. In like manner the streptococcus of erysipelas, in the event of its having been orignally present in the lymph material, is rapidly killed out by the germicidal action of the glycerin.

6. The tubercle bacillus is effectually destroyed even when large quantities of virulent cultures have been purposely added to the lymph.

7. The possibility of inoculation of syphilis is eliminated, as the calf is not subject to this disease.

8. The necessity for collecting children together, with the attendant risk of spread of infectious diseases, or of transporting a calf from place to place, is obviated, while the danger of "late" erysipelas in the child is diminished by reason of there being no necessity to open the mature vesicles for the purpose of obtaining lymph.

9. The bacteriological purity and clinical activity of large quantities of lymph can be readily tested prior to distribution.

10. By reason of the possibility of keeping large stocks of glycerinated lymph on hand for considerable periods of time without appreciable deterioration, any sudden demand, such as is likely to arise on the outbreak of epidemic smallpox, can be promptly met.

11. The expense of producing glycerinated lymph is proportionally small, since the amount obtainable from each calf is enormously increased.

With reference to the question of quantity Dr. Copeman further observes: "It is no unusual thing abroad to provide from a single calf an amount of glycerinated lymph that shall serve for four thousand and six thousand vaccinations, and in Berlin we were assured that the glycerinated lymph which was prepared in our presence from one calf, would suffice for no less than fifteen thousand vaccinations" (p. 192).[12]

Upon the subject of the duration of activity of glycerinated lymph Dr. Copeman also states: "Glycerinated calf lymph can be prepared which becomes freed from extraneous organisms, is available for a large number of vaccinations, at least five thousand from an average calf, and retains full activity for eight months, and will, under favorable circumstances, continue to do so in all probability for still longer periods if necessary" (p. 186).

Description of the Vaccine Establishment at Cologne.—This new vaccine plant, erected about 1895, was built as a matter of convenience near the new cattle and slaughtering establishment of Cologne.

In building the new establishment the provision of a suitable annex for the production of lymph was borne in mind from the very first; and so the present institution originated, which meets all the demands of hygiene, and may justly be regarded as the prototype of such institutions.

The building is very solidly built, but undercellared only, and to the smallest extent on the eastern side. As, however, the whole of the ground was filled in, and only absolutely dry and permeable material employed, there is no fear of any dampness of the rooms.

On the ground floor of the building are the corridor, collecting-room, calf stable, slaughter-room, doctor's room, office, and a closet.

On the upper floor, which is reached from the collecting-room by a convenient winding staircase, is the laboratory, adjoining which, on both sides, is a large garret. The calves are driven into the station through the doorway

of the slaughtering-room, which lies at the opposite end of the building. (See Fig. 4938.)

While the floors of the slaughtering-room, calf stable, collecting room, and laboratory are of concrete, the doctor's room and the office have inlaid wooden floors (parquetry); the corridor and closets are laid with "Mettlacher" tiles; and, finally, the rooms in the roof (garrets at the sides of the laboratory) are laid with floor boards. In the calf stable, slaughtering-room, and collecting-room the floor is slightly sloped in one direction, and at the lowest point there is a drainage outlet which is shut off by a small intercepting trap. The collecting-room, calf stable, and closet are lined, to the height of 1.5 metres from the floor, on all four sides, with white opaque glass tiles; the slaughtering-room on three sides (the door side is excluded) to the same height with white glazed tiles. The remaining part of the walls of these rooms is painted with white porcelain enamel. The walls in the doctor's room and the office are papered. In the laboratory and corridor the walls are painted with oil paint. The collecting-room is brightly lighted by a window 4 metres wide by 3 metres high. The light entering becomes strongly reflected by the brilliant white walls. The laboratory has three windows, the calf stable two, and the doctor's room and office one large window each. The closet and slaughtering-room have each one small window. All the rooms are, therefore, amply lighted.

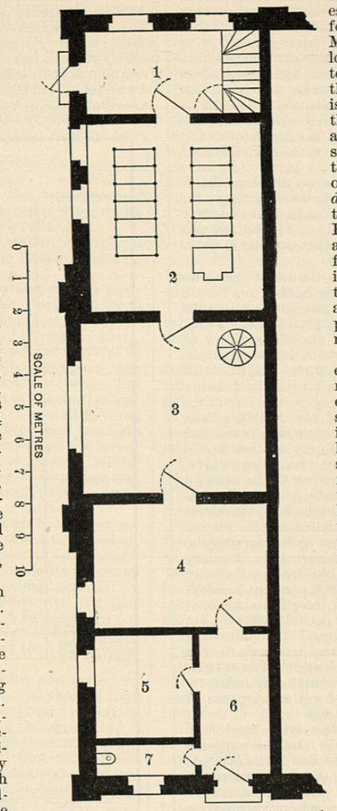

Fig. 4938.—Plan of Cologne Vaccine Station. 1, Slaughtering-room; 2, calf stable; 3, vaccinating-room; 4, medical director's room; 5, clerk's room; 6, entrance hall; 7, closet. (From *Centralblatt für allgemeine Gesundheitspflege.*)

The calf stable contains eleven stalls for the reception of calves. Upon the space which would correspond to the twelfth stall a weigh-bridge is sunk, enclosed with the above-described iron work; this machine allows of the calf being weighed while being taken through. The hollow in which the weigh-bridge stands has also a smell preventing arrangement. The ventilation of the stable is obtained by a large tube which passes through the garret above. The doors, padding, and air draught isolate the collecting-room both from smell and from noise. Between the calf stable and the collecting-room there is an arrangement of double doors, one of which is thickly padded. Water is supplied by the city main, and in every room a sink is provided. The lighting is by electricity; in the laboratory, however, the "Auerches Glühlicht" (a form of incandescent light) has been found preferable.

The heating is effected by means of American stoves. Large gas stoves serve for the heating of the water and milk. For the storage of considerable quantities of lymph a sufficiently large room is reserved in the refrigerating house of the city cattle establishment.

The laboratory is completely fitted for bacteriological investigations, containing all sterilization apparatus—thermostats, an excellent microtome, microscopes, centrifuge, etc.

The extent of the lymph production may be understood from the following figures: In the year 1894 about 356,000 portions of lymph were issued, and in this year (1895) about 400,000.

Calves.—The calves range usually from about six to eight weeks old. They are purchased in the meat market which immediately adjoins the establishment. The calves required for the current week are bought on Monday, and they are sold on the following Saturday, after their slaughter and the collection of lymph on the previous day; so that the stable is always empty from Saturday to the following Monday. The calves are kept under observation in the stable for twenty-four hours after their reception, and are vaccinated on Tuesday. *Only three complete days* are allowed for the progress of the local results of vaccination. On Friday the animal is slaughtered in a small slaughter-house opening off from the stable, and immediately it is dead the carcass is brought into the collection-room on a trolley, the abdomen washed, and the epithelial pulp of the vaccination area is removed by means of a sharp spoon.

Vaccination of Calves.—The process of vaccinating the calves is carried out in a fashion similar to that employed at the Berlin and Dresden stations, namely, by long parallel incisions over which glycerinated lymph is rubbed by means of a spatula or other flat-bladed instrument. The lymph employed for vaccination of calves is always kept for a period of at last six weeks after glycerination, in order to insure that it shall be as free as possible from extraneous organisms before it is used to vaccinate the calves.

After collection of the lymph pulp, and while it is being prepared for use, the carcass of the calf is taken back to the slaughter-room, where it is skinned and opened. The internal organs are removed and brought in on trays to be examined by the veterinary surgeon. In the event of his forming an opinion that any of the organs presented any condition indicative of disease, the lymph derived from the animal in question would be at once destroyed.

In view of this precautionary measure it is not deemed necessary to test the calves by the injection of tuberculin prior to their vaccination.

Collection and Preparation of Lymph.—The greatest amount of vaccine is collected during the months of March, April, and May, when from six to eight calves are employed every week. For the remainder of the year the weekly vaccination of one or two calves is found to be sufficient to supply all the lymph required for human vaccination and revaccinations in the Cologne district.

In the preparation of lymph material the epithelial pulp from the vaccinated area is removed by scraping with a Volkmann's spoon, and is received in a small glass dish. In this it is weighed, after which it is turned out into a mortar and thoroughly triturated; at first, without any addition of water or glycerin; later, small quantities of water are gradually added to the extent of five times the weight of pulp. The mixture having been

ground up still further, double the quantity of glycerin is finally incorporated. Thus, at the time of our visit, 10 gm. of pulp having been collected from one calf, the composition of the finished emulsion was as follows: Pulp, 10 gm.; water, 33.5 gm.; glycerin, 66.5 gm. Dr. Vanselow informed us that this quantity would suffice for the vaccination of five thousand children.

FIG. 4939.—Volkmann's Spoon.

The emulsion is afterward forced into small bottles and tubes by means of a machine actuated by water power. This machine, which is manufactured by a Vienna firm, appears to be decidedly useful and convenient, and is capable of being worked in connection with any form of pressure apparatus.

The small bottles are of different sizes, and contain lymph sufficient for fifty, for one hundred, and for one hundred and fifty vaccinations respectively. The bottles and their corks are all of them sterilized prior to use.

We were struck with the numerous precautions which are taken in this institution to insure thorough asepsis throughout the various stages of lymph production. Thus, in the collecting-room the flooring is of cement concrete, the walls are lined internally for about half their height with opaque glass tiles, the upper half being of Parian cement.

On the first floor of the building is a bacteriological laboratory, which is reached from the collecting-room by an iron spiral staircase. It is fitted up with an autoclave, incubators, etc. But owing to the director being engaged in private practice, he has no time to work in this laboratory, which is, therefore, used only when it is desired to sterilize cloths, instruments, or glassware. With the lymph and pulp material collected and glycerinated in our presence on January 16th, 1897, Dr. Cory vaccinated 34 children on January 19th, with an insertion success of 98.8 per cent., and 21 children on February 11th, with an insertion success of 93.3 per cent. (Copeman's Lectures, pp. 225–228).

Plan of Model Vaccine Establishment.—The following plan of a model vaccine establishment is shown in Delobel and Cozette's "Vaccine et Vaccination," Paris, 1899. The general plan is that of an open court surrounded by apartments, stables, etc., suitable for the purposes of producing vaccine material from animals.

This establishment is about 77 feet wide by 155 in length, and may be made much smaller by the omission of the two large director's rooms (intended in the original plan as a living-room) and the horse stable and carriage-house, as circumstances may require. Rooms 8 and 9 are found to be convenient in those cities where vaccination with fresh lymph is practised at the establishment as one of the functions of such a plant, a practice which was carried out for many years at the government station managed by Dr. Cory in London. (Fig. 4940.)

THE BACTERIOLOGY OF VACCINIA.—As early as 1809 Sacco[16] noticed the presence of granules in vaccine lymph. For a half century no further progress appears to have been made. As late as 1863 Eiselt of Prague described vaccine lymph as a contagious fluid devoid of cells. In 1866 Beale[17] drew attention to particles of germinal or living matter invading the tissues from the blood, which he identified as the virus contagium, or *materies morbi* of contagious diseases, and applied this principle especially to vaccine lymph. Hallier and Zurn,[18] of Jena, claim priority, and described in 1867 an abundance of micrococci as occurring in vaccine lymph. They also indicated the necessity for cultivation experiments to determine whether or no these micrococci stand in a causal relation to vaccinia.

Keber,[19] of Danzig, in 1868 also described micrococci in vaccine lymph. Cohn[20] in 1872 confirmed Keber's observations and distinguished the micrococci from the spores of moulds, and also differentiated them from bacilli. Cohn was not able to establish a difference between vaccine and variolous lymph.

It then became necessary to determine whether this particulate matter was essential to its specific action. Chauveau[21] attacked this question, and showed that the leucocytes in lymph are not essential to its specific action, since, after dilution, the supernatant fluid, free from leucocytes, was found to produce normal vesicles of vaccinia. On the other hand, by a carefully conducted experiment, he proved that if distilled water be cautiously

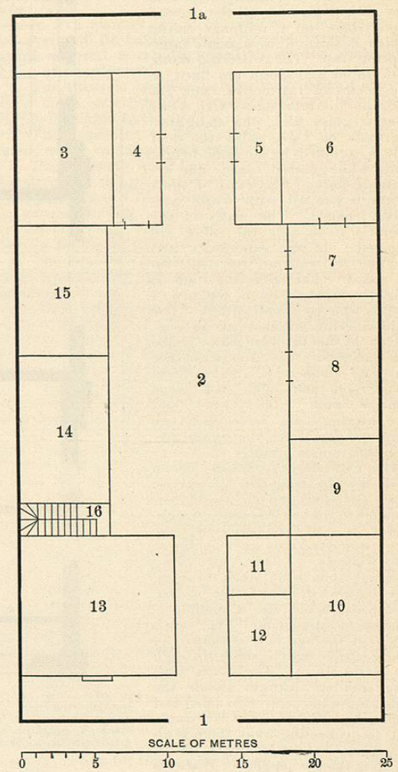

FIG. 4940.—Plan of Proposed Plant for Animal Vaccine Production. 1, 1a, Gates, one at each end. 1, Public entrance; 1a, entrance for animals; 2, open court; 3, stable for ten animals; 4, room for preparing the calves, shaving, etc., sufficient for one or two animals; 5, vaccinating-room, provided with one or two vaccinating-tables; 6, room for the vaccinated animals; 7, room for collecting the lymph; 8, vaccinating-room, for those who come to the establishment to be vaccinated; 9, waiting room; 10, laboratory; 11, packing-room; 12, clerical room, records and correspondence; 13, director's room; 14, carriage house; 15, horse stable; 16, stairway leading to the granary and room of employees.

floated on the surface of lymph in a small cylindrical vessel the soluble elements of the plasma will diffuse in the water, but the granular corpuscles remain in the lymph. The upper layers of the liquid, which have imbibed by diffusion the soluble albuminous matter of the lymph, are nevertheless totally inactive and incapable of

producing vaccinia. He therefore concluded that the active principle of the lymph resides, neither in the leucocytes nor in the soluble albuminous material, but in the suspended granular corpuscles of the lymph, that is, that the contagium is particulate. He showed further that lymph can be greatly diluted, and yet retain the contagium unimpaired, provided the greater the dilution the more extensive the absorbing surface. With a lymph diluted with about fifty volumes of neutral fluid a large proportion of vaccinations may fail totally; but in those which "take" the vesicles will not show any sign of attenuation or diminished virulence of the infective matter, but will be as large and as characteristic as those produced by undiluted lymph.

Sanderson[22] in 1870, in his report to the Privy Council, confirmed the experiment of Chauveau.

Copeman[16] has recently performed the experiment of filtering lymph through a Kitasato porcelain filter under pressure. The filtrate was absolutely sterile.

The hypothesis, therefore, that the active principle of vaccine lymph resides in its particulate elements rests on a very high degree of probability.

The flora of the lymph of vaccinia and variola have been thoroughly studied by Crookshank, Copeman, Blaxall, Buist, Fremlin, and others in England, by Cohn, Kleps, Koch, Pfeiffer in Germany, and by many others, and many micro-organisms have been isolated, several of which have been claimed to be specifically pathogenic, but no organism appears to have been discovered which has been proven to be the specific contagium of vaccine lymph. Of the various micro-organisms that have been discovered in lymph by different observers, the three most common are *Staphylococcus albus*, *S. pyogenes aureus*, and *S. cereus flavus*, which probably correspond to Buist's white, orange, and yellow vaccine. According to Copeman, one or another of these is to be found almost universally in every specimen of lymph examined.

Parallel with and in harmony with these investigations are the various experiments which have led to the adoption of glycerin for the purpose of preserving and diluting vaccine lymph, and of rendering it sterile, so far as concerns the presence of aërobic extraneous bacteria.

Cheyne[14] in 1850 and 1853 first demonstrated the fact that the activity of vaccine-lymph could be preserved for at least six months by the use of glycerin, and that its activity appeared to be increased rather than diminished by this means.

Müller,[23] director of the Berlin vaccine institution in 1866, confirmed Cheyne's experiments, and showed that such dilutions could be transmitted to distant countries and tropical climates without impairing their activity.

Copeman[24] again confirmed these experiments in 1891. In 1892 Chambon and Menard[15] found that while fresh glycerinated vaccine pulp gave only moderate results, the same emulsion kept for a fortnight caused a fair eruption, and at the end of forty, fifty, or sixty days produced a crop of typical vesicles.

Copeman found that a sample of glycerinated calf lymph forty-two weeks old gave in Dr. Cory's hands an insertion success, based on fifty insertions, of one hundred per cent.

Farrar[16] therefore concludes that prolonged glycerination secures the gradual extinction of all extraneous saprophytic microbes, that whatever be the nature of the contagium of vaccinia it is not an organism which can be cultivated by aërobic methods in ordinary laboratory media, and that no microbe isolated down to the year 1892 had any claim to be considered specific.

Crookshank[24] indorses this opinion, and says that no evidence is to be obtained by present methods as to the bacterial nature of calf lymph. He therefore suggests that a different line of inquiry may throw more light on the subject.

Other Theories.—Protozoa.—Renaut[25] in 1881 noticed certain highly refractile bodies in vesicles produced by inoculating the corneæ of rabbits and guinea-pigs with vaccine lymph.

Guarnieri[26] in 1892 described these bodies and called them *Cytoryctes vaccinia vel variolæ*, classified them as Rhizopoda, and claimed for them a specific action in vaccinia and variola. They are capable of reproduction. Monti,[26] of Pavia, identified the same bodies, and drew the same conclusions. Ruffer and Plimmer[27] independently examined the same bodies and established a close correspondence between them. The Pfeiffers,[28] of Jena, and Weimar,[29] Clarke,[30] and others also described these so-called parasites more carefully, confirming Guarnieri's hypothesis. It cannot be denied that by their delicate and careful work, they have made out a strong *prima-facie* case for the protozoon theory, which also derives much support from the analogy of malaria. But the malarial parasite is much more definite in shape and size, its developmental changes follow a regular and well-defined cycle, and the evidence of its being the specific etiological agent of malaria has been well established.

Ferroni and Massari[31] maintained, *per contra*, that bodies indistinguishable from the so-called parasites of Guarnieri could be identified in the epithelial cells after mere mechanical or chemical irritation, and that those bodies are either altered leucocytes or derivatives of the nuclei of the epithelial cells.

Salmon[32] examined these alleged protozoa and pronounced them to be merely chromatic masses derived from the migratory cells.

Attempts to cultivate protozoa have not met with success. Shattock and Ballance[33] in 1895 made many careful attempts at cultivation, but the results of all their experiments were negative. In 1892 a fresh impetus was given to the bacterial hypothesis by Klein,[34] who suggested that a bacterial organism might exist in vaccine and in variolous lymph latent in spore form.

Certain considerations favor this hypothesis: First. The action of glycerin appears to be germicidal only for cocci and sporeless bacilli, and is not inimical to the preservation of the spores.

Second. Lymph when dried after its collection from the vesicle preserves its efficacy for a long time, as well as or better than pure lymph stored in the fluid state. This fact suggests that active vaccine matter exists in the form of spores, since non-spore-bearing bacteria rarely retain their vitality when preserved for a long time in a thoroughly dry state.

Third. Lymph taken from the calf on the fifth, sixth, or seventh day, or from the human arm on the seventh, eighth, or ninth day, that is, when the vesicles are mature, retains its efficacy better than lymph taken at an earlier date, although such earlier lymph used directly from calf to calf or calf to arm is fully efficacious.

These facts are consistent with the existence of spores in the lymph taken at that particular date, and a microbe yielding spores as the last phase of its life history would be present in the earlier stages of vaccinia chiefly as a non-spore-bearing form, and in such form would withstand drying less well.

The earlier, therefore, we obtain lymph from the nascent vesicle the more likely we are to obtain the hypothetical microbe free from spores. By a special technique Klein succeeded in isolating a small bacillus, staining with difficulty, which there is fair ground for presuming to be the pathogenic organism of vaccinia.

Klein's method consists in prolonged staining with alcoholic gentian violet, by which he isolates a bacillus which he describes as occurring in cylindrical rods, fairly uniform in length, single, and occasionally in dumb-bells, 0.4 μ–0.8 μ in length, and one-third of this measurement in thickness. They are generally thicker at the ends than in the middle, and often include a spherical and slightly oval clear globule. They are to be found sparingly in lymph of the fifth or sixth day, more abundantly in that of the fourth day, and almost as a pure culture in lymph of the third day. Preparations of lymph taken from the calf seventy-two hours after vaccination contain bacilli chiefly in groups which look as though they came from an artificial culture. The later the lymph is taken the less numerous are the bacilli found. Klein made many attempts to cultivate this

bacillus in artificial media, but all attempts failed, This proves that the bacilli though constant in the lymph do not belong to the saprophytes that are occasionally present in lymph and are easily cultivable. Until this microbe can with certainty be cultivated, it cannot be submitted to the test of Koch's postulates, and its pathogenicity cannot, of course, be conclusively proven. Meanwhile, it would appear that there is a high degree of probability that it will turn out to be the specific agent in variola and vaccinia. This bacillus was also found in clear lymph from the vesicles of confluent smallpox of the fourth day.

Copeman claims to have cultivated the same or a similar bacillus by incubating for a long period—about a month—in eggs, and by inoculation of this upon calves he obtained a strain of lymph which was successfully used in the vaccination of calves and afterward of children.

After Chauveau had demonstrated that subcutaneous injection of vaccine lymph into the tissues is protective against subsequent vaccination (vaccinia sine exanthemate) Beclere, Chambon, and Menard[35] in 1896 proved that the serum of calves rendered immune by vaccination, and withdrawn several days or weeks after desiccation of the pustules, and inoculated into other calves in sufficient quantities (say 1,500 c.c.), renders these calves, if not absolutely, at any rate relatively, immune in a high degree to vaccination with lymph of proved potency.

Finally, Dr. Farrar concludes that the pathogenic identity of vaccinia and variola has been proved by the experiments of several workers. Klein's work on this subject is convincing. He showed that lymph originally derived from the vesicles of human variola and transmitted through calves would produce typical vaccine vesicles in calves and typical vaccinia in the human subject, the result being constant through several removes, and the calves so treated being insusceptible to vaccination with current calf lymph.

In the discussion which followed the reading of Dr. Farrar's paper Tavel,[16] of Berne, expressed the opinion that of all organisms hitherto found Guarnieri's *cytoryctes* would most probably be shown to be the specific etiological agent in vaccinia. He believed that the alleged discovery of a *sporidium vaccinale* by Funck, and the finding of certain protozoa by other experimenters were due to erroneous methods.

A new stimulus was given to the study of this most interesting question by the offer of a prize of £1,000 by the Worshipful Company of Grocers of London, in 1888. The problem being stated as follows: "To discover a method by which the vaccine contagion may be cultivated apart from the animal body in some medium or media not otherwise zymotic; the method to be such that the contagion may, by means of it, be multiplied to an indefinite extent in successive generations, and that the product, after any number of such generations, shall (so far as can within the time be tested) prove itself of identical potency with standard vaccine lymph."

The great importance of such an inquiry can scarcely be overestimated, since certain great advantages would thus be secured. On the one hand, the ability to produce unlimited quantities of vaccine material for immediate use in the face of an epidemic of smallpox, and, on the other hand, the ability to overcome the objections urged against animal virus of either sort, humanized or non-humanized, namely, the possibility of transmission of disease, however slight the foundation of such objections might be.

Experiments were conducted in this direction by Quist of Finland, by Dougall, and by S. C. Martin of Boston, but no practical conclusions appear to have been reached.

In harmony with this line of investigation are the investigations of Drs. Councilman, Magrath, Brinckerhoff, and Tyzzer. At a meeting of the Boston Society of Medical Sciences. April 28th, 1903, Dr. Councilman announced the discovery of the etiology of smallpox, and at the same time gave his opinion as to the identity of the disease with vaccinia.

He stated that smallpox passes though certain perfectly definite stages in its development on the clinical side, to which correspond equally definite changes in the form of the causative organism; that these changes in form consist first in the appearance in the cell protoplasm of small, homogeneously staining, structureless bodies, about 1 μ in diameter, not staining so deeply as nuclei, which gradually increase in size, coincidently with the degeneration of the cells in which they lie; that with this increase in size the character of these bodies changes, small granules surrounded by circles appear in them, the general outline becomes irregular, with increasing resemblance to an amœba; that a process resembling sporulation takes place with the breaking up of the amœboid forms into smaller, sporelike bodies; and that this constitutes the first cycle in the life of the organism. Up to this period the nuclei of the epithelial cells show no changes whatever, but the following alterations in these structures now appear: penetration of nuclei by the small sporelike bodies before spoken of, with the disappearance of such bodies from the cell protoplasm; small ring-shaped bodies become apparent in the nuclei; they also increase in size; the ring shape is at first preserved, which is soon surrounded by a spongelike body. This whole structure grows, fills, and finally destroys the nuclei, and the bodies become free; the bodies again become more homogeneous; ring-shaped bodies again become evident; finally each ring appears to have a central dot. These final bodies are regarded as the infecting agent of the disease. Two cycles of the organism, corresponding in general to what we know of the protozoa, have therefore been demonstrated: the first—extranuclear—simple, and the second—intranuclear—very probably sexual.

It also appears to be demonstrated by this work that vaccinia represents the extranuclear phase of the organism, but that the production of true smallpox is dependent upon the invasion of the nuclei—the intranuclear cycle. Vaccinia may be produced by inoculation, for example, in the calf or the rabbit; variola under similar conditions is produced in the monkey. This, also, Dr. Councilman and his colleagues have shown. The organism was found in the blood in one case. The difficulties of study are very greatly increased by the extreme rapidity with which the entire process takes place and by the ease with which the organisms are destroyed; the life history is completed with the formation of the vesicle, hence the absolute necessity of securing early and perfectly fresh material. The rôle of bacteria is regarded as entirely subordinate to that of the organism described, though the influence, particularly of streptococci, must be taken into consideration in the development of the disease.

Dr. Councilman expressed the opinion with confidence that the organism which he described as belonging to the protozoa was the essential etiological factor in the production of smallpox.

Proposed Investigations Relative to Vaccination.—The offer of a prize by the Grocers' Guild of London amounting to £1,000 has already been referred to, the object of this prize being to stimulate investigation in the direction of some possible medium of vaccine culture outside the living organism, either of man or of the lower animals.

A new line of investigation is also suggested by the Report of the German Board of Health of 1900, in which it is stated by the vaccine authorities of Hamburg, that they have reason to believe from the experience which they have had in the culture of vaccine lymph that it is possible to secure a much longer period of immunity from smallpox than has hitherto been possible by the employment of any vaccine lymph in ordinary use. They had arrived at this conclusion in consequence of certain observations relative to the percentage of failures in the revaccination of school-children in that city. It was observed that, for some reason, in 1894 the percentage of successful vaccinations among the revaccinated children in Hamburg suddenly fell off from an average of about 85 per cent. in the nine years 1885–93 to an average of only 67 per cent. in the six following years,

1894–99. That this decline in the percentage of successful revaccinations was not due to any deterioration of the lymph in use appears to have been proven by the fact that the high percentage of success among primary vaccinations conducted with the same lymph (an average of fully 98.5 to 99.5 per cent.) continued throughout the entire period. The only explanation offered for this fact was that in consequence of employing a lymph of unusual strength in the earlier primary vaccinations, these children were rendered immune for a longer period than they would have been had ordinary lymph been employed, so that when they arrived at the legal age for revaccination the percentage of successful results was greatly diminished.[8]

This fact opens up a very interesting field of investigation with the view of strengthening the quality of vaccine lymph to such a degree as to prolong its immunizing power.

The following figures illustrate this singular fact in the experience of the Hamburg vaccine district:

RESULTS OF VACCINATION AND REVACCINATION IN HAMBURG DURING THE YEARS 1885–1901.—PERCENTAGE OF SUCCESSFUL RESULTS.

Years.	Primary vaccinations.	Revaccinations.	Years.	Primary vaccinations.	Revaccinations.
1885.....	99.8	85.8	1894.....	98.8	69.3
1886.....	95.4	78.9	1895.....	99.7	66.5
1887.....	98.9	80.2	1896.....	99.3	59.7
1888.....	99.8	83.4	1897.....	99.4	66.8
1889.....	99.8	87.7	1898.....	99.5	69.6
1890.....	99.7	90.1	1899.....	97.7	69.5
1891.....	99.4	89.3	1900.....	99.4	74.8
1892.....	99.0	87.3	1901.....	99.7	93.6
1893.....	99.6	82.9			

LAWS OF DIFFERENT COUNTRIES RELATIVE TO VACCINATION.—Herewith are presented the essential features of the vaccination laws of different countries in which vaccination has been introduced.

England.—The following are the essential provisions of the vaccination act of England as amended by further acts of 1871, 1874, and 1898:

(I) 1. The period within which the parent or other person having the custody of a child shall cause the child to be vaccinated shall be six months from the birth of the child, instead of the period of three months mentioned in § 16 of the vaccination act of 1867, and so much of that section as requires the child to be taken to a public vaccinator to be vaccinated shall be repealed.

2. The public vaccinator of the district shall, if the parent or other person having the custody of the child so requires, visit the home of the child for the purpose of vaccinating the child.

3. If a child is not vaccinated within four months after its birth, the public vaccinator of the district, after at least twenty-four hours' notice to the parent, shall visit the home of the child, and shall offer to vaccinate the child with glycerinated calf lymph, or such other lymph as may be issued by the local government board.

4. The public vaccinator shall not vaccinate a child if, in his opinion, the condition of the house in which it resides is such, or there is or has been such a recent prevalence of infectious disease in the district, that it cannot be safely vaccinated, and in that case shall give a certificate (under § 18 of the vaccination act of 1867) of postponement of vaccination, and shall forthwith give notice of any such certificate to the medical officer of health for the district.

5. Notwithstanding any regulation of any lying-in-hospital or infirmary, or other similar institution, the parent of any child born in any institution shall not be compelled under such regulation or otherwise to cause or permit the child to be vaccinated at any time earlier than the expiration of six months from its birth.

(II) 1. No parent or other person shall be liable to any penalty under § 29 or § 31 of the vaccination act of 1867, if within four months from the birth of the child he *satisfies two justices*, or a stipendiary, or metropolitan police magistrate, in petty sessions, *that he conscientiously believes that vaccination would be prejudicial to the child*, and within seven days thereafter delivers to the vaccination officer for the district a certificate by such justices or magistrate, of such conscientious objection.

2. This section shall come into operation on the passing of this act, but in its application to a child born before the passing of this act there shall be substituted for the period of four months from the birth of the child the period of four months from the passing of this act.

3. An order under § 31 of the vaccination act of 1867, directing that a child be vaccinated, shall not be made on any person who has previously been convicted of noncompliance with a similar order relating to the same child.

4. No proceedings under § 31 of the vaccination act of 1867 shall be taken against any parent or person who has been convicted under § 29 of the said act on account of the same child, until it has reached the age of four years.

5. Persons committed to prison on account of noncompliance with any order or non-payment of fines or costs under the vaccination acts shall be treated in the same way as first-class misdemeanants.

Italy.—Italy supports fourteen vaccine establishments, at Bologna, Milan, Genoa, Ancona, Venice (two), Bergamo, Arezzo, Verona, Vicenza, Rome, Modena, Ravenna, and Rimini.

A law of January 1st, 1892, provides for the production and preservation of vaccine lymph.

All children must now be vaccinated within the half-year succeeding their birth. Certain exemptions are specified—children who have had smallpox, etc.

Persons who have not been revaccinated since they were eight years old are excluded from schools and workshops until they are revaccinated.

Norway.—In Norway, vaccination was made obligatory by law in 1811.

In *Austria*, *Roumania*, *Turkey*, and in *Greece* the Government assumes the right to vaccinate all the unvaccinated, and in some of these countries to revaccinate all who have not been vaccinated within seven years. Vaccination is not compulsory in *Belgium* and in *Spain*.

In Vienna, Dr. Weyl attributes the low mortality from smallpox in the last years of the century to the introduction of the vaccination of school-children, to the active efforts of officials to arouse an interest in public vaccination, to the introduction of animal vaccine lymph, and to the establishment of a state institution for this purpose. ("Die Assanierung von Wien," 1902, p. 173.)

France.—After a century of failures to enact a compulsory vaccination law France has finally fallen into line by the enactment of a law which became operative in February, 1903. This act constitutes a portion of the new "law for the protection of the public health" of February 19th, 1902. It provides: (§ 6) that vaccination shall be obligatory during the first year of life, and also that revaccination shall be performed during the eleventh and twenty-first years. Parents and guardians are made responsible for the execution of this measure. A public regulation made after consultation with the Academy of Medicine and the Consulting Committee of Public Health will establish the necessary measures for carrying out the provisions of this act. By Article 23 the prefect of police is charged with the duty of enforcing the provisions of the act so far as penalties are concerned.[36]

Vaccination of all new recruits in the French army is obligatory, and to this regulation is undoubtedly due the comparative immunity of the army from smallpox as compared with the people at large.

Holland.—In Holland vaccination is not compulsory, and for many years it was the custom to delay vaccination until the child was more than two years of age.

There are at present four vaccine establishments, at Rotterdam, Amsterdam, The Hague, and Utrecht, and three temporary establishments at other places. Bovine lymph is mainly used at these.