THE CROONIAN LECTURES ON THE CHEMICAL PRODUCTS OF PATHOGENIC BACTERIA CONSIDERED WITH SPECIAL REFERENCE TO ENTERIC FEVER.

Delivered before the Royal College of Physicians of London,

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LECTURE I.

I PROPOSE in the following lectures to discuss generally the nature and action of the poisons and chemical products formed by pathogenic bacteria, and to illustrate the subject by an investigation that has occupied me recently of the pathogenic action and the chemical pathology of the typhoid bacillus and of two other bacilli closely related to it.

Most of the poisons which are to be considered are substances which cannot be defined chemically, and this at the outset is a very great difficulty in an attempt to classify such poisons accurately.

The transformations which bacteria, both non-pathogenic and pathogenic, produce in substances are of great variety. Some non-pathogenic bacteria are concerned, for example, with nitrification, a process in which ammonia compounds are transformed into nitrates and nitrites. These bacteria are more or less specific, and although some pathogenic organisms have a somewhat similar action in this respect it is very slight. Another example of a similar character is the splitting up of urea into ammonia and carbonic acid by the micrococcus ureæ. These actions, however, are not of the nature of the chemical changes produced by pathogenic bacteria. The changes which bacteria produce in carbohydrate substances—for example, starches and sugars—are, first, the transformation of the starch into one or other kind of sugar, and the decomposition of the sugar into alcohol and acid bodies, such as lactic, acetic, and butyric acids. Such changes are usually spoken of as those of fermentation, and are allied to changes which occur in the digestive processes of the higher animals by means of the organic as distinguished from the organised ferments.

Some bacteria are capable of still further acting on the acids produced in the first-named fermentations; thus the bacillus butyricus can transform lactic acid into butyric acid with the formation of carbonic acid and hydrogen. These are again not the changes which are of the greatest importance in considering pathogenic bacteria. Although some pathogenic bacteria act on starches and sugars with the formation of acid bodies and gases, yet the products so formed play but a small part in the production of disease. Fats, which are largely present in the body and are taken in with food, are also broken up by bacteria of one kind or another. The products, however, from a pathological point of view, are of minor importance. From the albuminous substances, however, the third great class of organic substances that serve as food and act as important chemical constituents of the body, the pathogenic bacteria produce substances possessing important physiological actions. The albuminous molecule containing carbon, hydrogen, nitrogen, sulphur, and frequently phosphorus, is unknown as regards its chemical constitution. It is no doubt highly complex, and is capable of being broken up both by organic ferments and by bacteria into numerous bodies of the most complex nature. Although the actual constitution of the albuminous molecule is at present undetermined, it is capable of being transformed only in certain directions. Thus, by the action of mineral acids and alkalis with the aid of heat, white of egg or the serum proteids are transformed into acid and alkali albumin, which differ from the original proteid mainly in their solubilities in acid or alkaline solution. The acid form is precipitated by an alkali and *vice versa*, the precipitate being still capable of coagulation.

Like the original proteid, however, these bodies are capable of digestion. The digestive transformation which takes

place in the stomach and small intestines consists in the change of these slightly soluble and coagulable proteids into more soluble and diffusible ones, the formation of the so-called albumoses and peptone. The important members of this class are soluble in water, are not precipitated by heat, and are capable of some amount of diffusion. They all give a bright pink colour with a trace of copper sulphate solution and an excess of potash. True peptone differs from some of the albumoses in the fact that it is not precipitated by nitric acid. One of the characteristic albumose reactions, for example of proto-albumose, is that the precipitate by nitric acid is soluble on heating the solution, and comes down again on cooling.

Peptic digestion probably does not go farther than the formation of peptone, and results chiefly in the transformation of the proteids into albumoses. Pancreatic digestion, however, can split up the proteid molecule, or part of it, into crystalline bodies containing nitrogen, such, for example, as leucin and tyrosin.

This digestive action on proteids produced by organic ferments is also a feature of the action of some of the organised or bacterial ferments, except that the effect is more pronounced, and results in the breaking up of proteid molecule into many different non-proteid substances, which may be considered the final products of the digestive action.

This digestive action of pathogenic bacteria is, however, only a part of the question we shall have to consider. The chief cell of the peptic gland secretes pepsin, which is discharged in the gastric juice into the stomach cavity; the pancreatic cell secretes trypsin and other ferments, which are discharged into the small intestine with the pancreatic juice; and, therefore, in any digestive mixture obtained from the stomach or intestine there is present, besides the products of digestion, the ferment or the active agent in the transformation. In the case of bacteria growing in an artificial medium outside the body or developing in the body, there is—in addition to what digestive products may be formed—a living bacterial cell, and any secretion which it may produce; and it will be seen that the body of the bacterium as well as its secretion—whether this be of the nature of a ferment or not—are both of the highest importance in the consideration of diseased conditions and processes.

In bringing these aspects of bacterial action to your notice, it will be seen to what extent the products of their digestion of albuminous substances or the mere secretion of poisonous substances act in disease.

The study of putrefaction, which is produced by many different micro-organisms, has shown that—besides the production of albumoses as the early products of digestion—numerous chemical substances, or final products, are formed, some of which are not toxic, although many are highly poisonous.

It is not necessary to dwell on this part of the subject, except to say that the effect of these toxic bodies of putrefaction is: (1) that of intense local irritation, whether they are injected under the skin or introduced into the digestive tract; and (2) an action on the central nervous system, whereby, besides the production of muscular twitchings and clonic and tonic spasms, there is with many of them a tendency to the production of stupor and coma, ending in death. These chemical products of putrefaction, with perhaps one exception—that of mydaine—do not cause a rise of body temperature, so that the fever which is observed in cases of poisoning by putrefactive products, such as those occurring in some cases of food poisoning, are due to other products of the bacteria. At one time similar chemical products or animal alkaloids, as they are called, were supposed to play a large part in the processes of specific infective diseases. Thus Brieger isolated two bodies, tetanin and tetano-toxin, which were supposed to be the active agents in the production of the convulsive spasm of tetanus. He also isolated typho-toxin, a chemical body which was supposed to be an active poisonous agent in typhoid fever. The existence of these bodies has, however, not been confirmed by subsequent research. It does not follow, of course, that in all diseases such final products may not play a part in the production of the symptoms. As I have shown in anthrax, such a product is formed, and is one of the active poisons

in the disease. Of later years, especially as the result of the study of the chemical pathology of diphtheria and of tetanus, the most active poison which is produced by the micro-organism is not of the nature of an animal alkaloid, but a substance which cannot be defined chemically, but only by the investigation of its physiological action.

SEPARATION OF BACTERIAL POISONS.

The three kinds of poisons which have been indicated must be borne in mind, namely: (1) The poisons secreted by the bacterium itself; (2) the products of the digestive action of the bacterium, that is, the albumoses; and (3) the final non-proteid products, which may be provisionally called an animal alkaloid. The first two poisons are precipitated from solution by means of alcohol; generally speaking, the last class of poisons is soluble in alcohol. In the separation of the first two classes of poisons by means of alcohol great care has to be taken that they are not exposed for too long a time to the action of the reagent, so that their toxic properties may not be diminished. The prolonged action of alcohol, for example, on pepsin will diminish its activity; and so it is found that its prolonged action on the poisons of diphtheria and tetanus greatly diminish their activity. Any method, therefore, in which alcohol is employed must be a rapid one, the poison not being exposed to its action for longer than a few hours.

Another method which is employed in the separation of these poisons is precipitation by means of saturation of the liquid with neutral ammonium sulphate. By this means all the proteids are thrown down with the poisons, and may be collected and redissolved by means of dialysis. To my mind, however, this method is open to grave objections. Ammonium sulphate is itself poisonous, and it is extremely difficult to remove the last traces of the salt, except after prolonged dialysis in the presence of antiseptics, which may be hurtful to the poisons. During the process of dialysis the solution becomes greatly diluted owing to the absorption of water from outside the dialyser; so that there is at the end a very dilute solution of the poison, which has to be concentrated before it can be used. There is no advantage in this method, because although it gives rapid precipitation of the poison, yet the final product is not any purer than when alcohol is used as a precipitant. Other methods have been used by Brieger and others whereby the poisons have been precipitated by the salts of heavy metals, which are got rid of afterwards by various means. Even in this case we are not much nearer the chemical identification of the poisonous substances.

PREPARATION OF SECRETORY PRODUCTS OF THE BACTERIA.

The two best known examples of toxic secretory products of bacteria occur with the bacilli of diphtheria and of tetanus. They are produced by growing the bacillus in a medium made of sterilised extract of meat, to which 1 or 2 per cent. of commercial peptone and a small proportion of common salt are added. This is the ordinary broth peptone. It is a medium in which the proteids exist in a digested form, so that any digestion of the proteids by the bacterium does not take place, as it might do if serum albumin or white of egg were present.

After growing in the medium for a certain time—three weeks, a month, or longer, and after filtering off the bacilli through a porcelain filter, a clear sterile liquid is obtained which is extremely poisonous. This liquid when injected into an animal gives rise, in the case of diphtheria, to the characteristic symptoms of the disease—namely, a palsy which is mainly dependent on the degeneration of the peripheral nerves; and in the case of tetanus to the characteristic symptoms of the disease. In this poisonous liquid no manipulation has as yet demonstrated the chemical identity of the poison. Such a liquid gives a copious precipitate with alcohol or saturation with ammonium sulphate, and this precipitate is itself highly poisonous, containing most of the poison present in the liquid, and consists almost entirely of the peptone which has been added to the liquid in the preparation of the culture medium with some of the salts of the broth.

An attempt was made by Roux and Yersin to isolate this poison by adding phosphoric acid and then lime to the

liquid, the precipitate of calcium phosphate formed carrying down the poison. It was found that the small amount of this calcium phosphate mixture which killed an animal did not show any appreciable difference in weight from the amount of calcium phosphate present—that is, the amount of poison present was practically imponderable. This may also be appreciated by the statement that even as little as $\frac{1}{10}$ c.cm. of the broth filtrate may kill a guinea-pig weighing 300 g. in forty-eight hours.

Kitasato also found in his experiments on the bacillus of tetanus that the broth filtrate was poisonous in what were practically infinitesimal doses. It is evident that in these cases we are dealing with a poison which is at present quite outside the domain of chemistry; it cannot be expressed chemically, it can only be expressed from its pathological effects.

SEPARATION OF THE DIGESTED PRODUCTS.

To obtain these it is necessary to grow the bacillus in a liquid containing either alkali-albumin, serum, or any of the liquids of the body containing proteids in solution. Extract of meat and salt (that is, broth without peptone) is with advantage added, as it thus makes a better culture medium for the bacterium. After the bacillus has grown in this liquid for three to five weeks, and it is filtered off through porcelain, the clear filtrate, after concentration at 40° C., is precipitated by an excess of alcohol, and the precipitate collected and redissolved in water, reprecipitated by alcohol, and after washing in alcohol and ether is dried over sulphuric acid. In this way a lightish brown powder is obtained, readily and completely soluble in water, and giving the reactions of albumoses—namely, the biuret reaction and the characteristic precipitate with nitric acid.

Some bacteria—for example, the anthrax bacillus—have practically no toxic secretion, and the albumoses are readily separated in as pure a form as can be obtained; but with others—for example, the diphtheria bacillus—where there is not only a secretory product, but also digested products, the powder which is obtained by the method thus stated contains not only the secretion, but also the albumoses. There are no chemical means of separating these secretory products from the albumoses, but some degree of separation may be obtained by keeping the solution at 60° C. for a longer or shorter period, whereby the secretory product is rendered inert. This is, of course, only a rough method, inasmuch as heat also damages, in some instances, the action of the albumoses.

SEPARATION OF THE FINAL PRODUCTS.

The third point is the separation of the final products. These final products are obtained in the alcoholic filtrate after precipitation of the proteids in solution. This alcoholic filtrate is evaporated to dryness at a temperature of 40° C., and extracted with absolute alcohol. The extract is filtered, evaporated again to dryness, and redissolved in alcohol. After filtration, and, if necessary, concentration, it is thrown into an excess of ether, which causes a precipitate, and the alcoholic extract may thus be separated into two parts, one of which consists of substances which are soluble in alcohol and insoluble in ether, and are precipitated by ether, and other substances—chiefly fats—which are soluble in ether and alcohol, but are insoluble in water.

This manipulation and separation of the substances is, of course, at present a very imperfect one, inasmuch as the substances so separated have not been obtained in a sufficiently large quantity, nor are their chemical characteristics sufficiently worked out to enable a definite idea of these characteristics to be obtained; but for the purposes of physiological experiment and of determination as to the general nature of the poisons produced by pathogenic bacteria, I have found this method of great value.

In the investigation of toxic products formed in infective disease, not only must they be studied by means of the artificial culture of the bacillus in suitable media, but a chemical examination of the body itself, especially as regards the blood, the spleen, and the liver, is necessary in order to determine whether the same products are found in the body as in the artificial culture.

This study is of great value when the poisons have a specific action; for example, in the case of diphtheria and of tetanus;

but in the diseases in which there is no specific symptom due to a chemical poison, it may be impossible to prove the identity of the poisons produced by the bacterium and those found in the bodies of patients dead from the disease.

INTRACELLULAR BACTERIAL POISONS.

The three classes of poisons just considered are described as extracellular bacterial poisons, but besides these, which are present in the culture fluid in which the bacteria are grown, poisons may be present in the bodies of the bacillus—intracellular poisons—and these have to be studied by means of other methods. One method is to kill the bacillus, which is grown in broth, by means of chloroform. If a few drops of chloroform are added to the liquid and shaken up, and allowed to act for twenty-four hours or longer, the bacilli are killed, so that, on testing, the liquid is found to be sterile. The bacilli are still suspended in the liquid, and after evaporation of the chloroform *in vacuo* the liquid may be used for experiment. Besides the bodies of the bacillus it of course contains any secretory poisonous product it may form. The results obtained by the injection of this liquid may be compared with those observed with a broth culture, also chloroformed, and from which the bodies of the bacillus are removed by centrifugalising. The comparison of the two results enables one to observe the difference in the action between the poison contained in the body of the bacillus and the secretory product; between the intracellular poison and the extracellular.

Another method consists, after growing the bacillus on a large surface of agar, in scraping the culture from the surface and suspending it in a sterile salt solution. Some amount of extracellular poison may be present in this suspension as well as in the first case. In filtering a culture fluid through a Chamberland filter some of the toxic products may be held back by the filter itself, and this is the more likely to be the case if the solution contains a large amount of undigested proteid, for example—serum. The method of killing the bacillus in the fluid by means of chloroform and centrifugalisation to get rid of the bodies of the bacillus is therefore a very important one in the investigation of the toxic products of the bacterium, as in this way the intracellular poisons of the bacillus can be investigated.

TOXIC ACTION AND REACTION.

The physiological action of these products may be investigated by studying the effects produced by their injection subcutaneously, into the peritoneal cavity, or into the venous system. As the result of the intravenous injection of poisons a more rapid effect is produced than by other methods, and observations may be made in rabbits as to the effect of the poison on the temperature, on the body weight and on the intestine (as regards the production of diarrhoea) as well as its effects on the production of microscopical lesions in the nervous system, the heart, liver, spleen, and kidneys.

From the experimental point of view infective diseases may be divided into two great classes. In one of these the symptoms are only those of what we must call a toxæmia or blood-poisoning—that is, there is a fever in a greater or less degree, with great bodily depression, and loss of weight, with some alteration in the secretion of the glands, more evidently those of the kidney and the skin. At some period or other of this process the fever may be absent, and yet the process of poisoning may be progressive. In the second class of cases, besides the symptoms of toxæmia, there are present during the acute disease, or following as sequelæ, specific lesions, mainly of the nervous system.

Examples of toxæmia without specific lesions occur in anthrax or malignant pustule, and in typhoid fever.

Examples of the second class are diphtheria and tetanus, in both of which, besides the toxæmia, there are specific symptoms referable to the nervous system.

The animals which have been used in the investigation are rodents (rabbits and guinea-pigs) between which and the higher animals there is a great gap, chiefly on the side of the nervous system. This point must be borne in mind in any deductions made upon the action of poisons upon the nervous system in rabbits.

The method of procedure in the experiments is as follows: The weight and rectal temperature of a healthy rabbit are taken for two days at least previous to the experiment. In this way an indication is obtained by the

increase of weight in the animal, and by the regularity of the temperature as to whether it is healthy or not. The injection is then made, usually into the marginal vein of the ear, and the effect of poison on the temperature, body weight, etc., observed during the life of the animal. After death, or at killing, cultivations are made from the heart's blood, the peritoneal cavity, and the spleen or liver, to demonstrate the absence of bacterial action, and the nerves (the phrenic and branches of the anterior crural) are stained with osmic acid for the investigation of degeneration, and a portion of the left ventricle of the heart is also stained in the same liquid for examination of any change in the fibres. Bacterial poisons appear to have a specific action on the heart of the rabbit in the production of degeneration of the muscle fibre, a degeneration which is the expression of the cardiac failure so frequently observed in infective disease in the human being.

One important point as regards the action of these bacterial poisons must be noted, namely, that they have in many cases a very slow action; so that following on a single dose there may be some slight change observed in the temperature or in the body weight, which passes off. Subsequently, the animal either progressively goes down hill, and succumbs after a week or fourteen days, or longer, or there may be no apparent downward progress, and death occurs suddenly.

This slow action of the bacterial products is so common as to be noted particularly, and in some cases it precludes any exact investigation of their effects on the circulation of the blood within the time limits of an experiment.

TOXIC REACTION.

Besides the poisonous effect of the bacterial products on the body whereby an injury is effected on the tissues, producing either alteration in function, with or without obvious degeneration of the tissue itself, bacterial products may produce what may be called a toxic reaction. This occurs most readily as the result of the injection into an animal refractory to the disease of small but gradually increasing doses of poison. There is no accumulation of the poison in the body, but there is a gradual increase in the blood and tissues of a substance or substances which are inimical to the poison or the bacillus—that is, under certain conditions they may prevent, when injected at the same time, the fatal effects of the poison on the bacillus.

These substances probably fall into two groups—(1) the antitoxic, which counteract the effects of the poison itself, and (2) the antimicrobial, which counteract the effects of the bacillus itself. In one and the same animal the blood may contain a substance or substances which are both antitoxic and antimicrobial, such, for example, as occurs in the process of the formation of the diphtheria and tetanus antitoxic serums.

As an introduction to the investigation which I have more recently done, it will be well to discuss as examples the chemical and pathological processes which occur in anthrax, diphtheria, and tetanus.

ANTHRAX.

I have previously shown, and subsequent experiment has only confirmed the observation, that the bacillus of anthrax produces in the body of an animal, the subject of anthrax, and in an artificial culture medium containing digestible proteids, a large quantity of albumoses, and also of a body which gives some of the reactions of an alkaloid. Both these classes of bodies are poisons, the albumoses producing fever and causing death in coma when given in large doses, the alkaloidal body, which is much more poisonous, producing no rise of body temperature, and causing stupor ending in coma and death in mice, guinea-pigs, and rabbits. According to the dose given death may not be produced, but the stupor and coma may last for a certain time and then be recovered from.

The bacillus of anthrax is a typical infective micro-organism, and it is the chief example of the class of micro-organisms whose toxic products are the results of the transformation of the proteid solutions in which it grows. Practically it produces no toxic secretion. If any is produced at all it must be in very small amount, inasmuch as when grown in peptone broth it imparts little or no toxicity to the liquid, as tested by injection after removal of the bacilli through the filter.

TETANUS.

This is quite different from the results obtained in the bacillus of tetanus. If this is grown in a solution of peptone broth under suitable conditions it is found to produce a highly toxic substance, which, as has been previously stated,

is practically unweighable. This substance, which must be considered a secretion of the bacillus, possesses a characteristic physiological action in that it produces the characteristic convulsions of the disease tetanus. It is the chief toxic agent produced by the bacillus. Kitasato once found it in the heart's blood of a patient dying of tetanus. In a number of cases of tetanus which I examined chemically I did not find this poison, although perhaps this result is not of much value, as the method I employed would possibly destroy the somewhat sensitive toxin. I found in the spleen and blood of patients dying of tetanus albumoses in fair quantity which did not possess any other action than that of producing fever; they produced no convulsions, and did not cause death. I have previously stated that in one experiment I found that the ether extract of the spleen in tetanus produced convulsions when injected intravenously in rabbits. This undoubtedly occurred, but I am not at all sure that it is of very great significance; at any rate, I could not confirm the observation, and I do not wish to lay any stress on it. It must be considered at present that the chief if not the only active poison in tetanus is the secretion of the bacillus, which, when injected into the body after a certain period produces convulsions.

With regard to the albumoses which are found in the body, in the spleen and blood of patients dying of tetanus, they are undoubtedly present. They may be in some cases the result of a mixed infection by the bacillus of tetanus and other bacteria, but they may be the result of the direct action of the tetanus bacillus or of its toxin, although the proof of this is at present wanting. I did not find that the tetanus poison had any digestive action.

DIPHTHERIA.

The chemical processes of diphtheria closely resemble those of tetanus. There is a secretory product which is formed when the bacillus is grown in peptone broth, and which is present in the membrane of diphtheria, and probably in the tissues as well. This secretory product possesses a characteristic action on the nervous system, causing in rabbits paralysis dependent on nervous degeneration. When the bacillus is grown in a solution containing digestible proteids, it digests these with the formation of albumoses and certain by-products. These albumoses, separated in the ordinary way, have a toxic action similar to, although not so powerful as, that shown by the secretory product. They are also found in the spleen and blood of persons dying of diphtheria, and possess a similar action. What part of the toxic action of the products is to be ascribed to the albumoses themselves is perhaps a matter of debate, inasmuch as the only method by which the physiological action of the secretory product can be separated from that of the albumoses is that of heat, which also affects the action of the albumoses.

What I have called the secretory products of tetanus and diphtheria are remarkably sensitive to heat; that of tetanus, for example, is completely destroyed by raising to a temperature below 80° C.; that of diphtheria is destroyed at a temperature of 60° C., and heating to lower temperatures weakens the action of the poison or alters its effects.

It is evident that the action of the powerful secretory products of pathogenic bacteria is unlike that of crystallisable poisons. First, they produce a large effect in very small doses; secondly, this effect does not always come on immediately after injection even into the venous system, but is preceded by a period during which no apparent change is occurring in the body; thirdly, the poisons are frequently selective in their action, affecting mainly the nervous system, on which they produce a profound effect. They may also disorganise the general nutrition of the body. Fourthly, they are very sensitive to heat and external conditions. Their injection into the body—their effect being preceded by a period of incubation—reproduces the action of the bacillus itself, so that one has to do with a poison which, although not living, produces a progressive change in the tissues of the body like the bacillus.

DR. ROBERT RIEDER, Extraordinary Professor of Surgery in the University of Bonn, has accepted an invitation from the Turkish Government to occupy the chair of surgery in the Military Medical Academy of Constantinople.

TWO LECTURES ON THE TEMPLES AND RITUAL OF ASKLEPIOS AT EPIDAUROS AND ATHENS.

Delivered at the Royal Institution of Great Britain

By RICHARD CATON, M.D., F.R.C.P.

LECTURE II.

TO-DAY we pass on to consider the ritual of the Asklepiian shrines and the accommodation and treatment of the sick who frequented them.

It is convenient, first, to consider the Hierarchy. This consisted of the Hieres or Hierophant, the priest, who was the head official. He was appointed annually, and he appears to have been frequently re-elected. From the Athenian inscriptions we know that sometimes he was a physician, sometimes not; so also it was with the subordinate officials. The priest was the general administrator, and had a share in the financial government of the temple. The Dadouchoi, or torch-bearers were probably subordinate priests; the Pyrophoroi, or fire carriers, among other functions, lighted the sacred fire on the altars; the Nakoroi or Zakoroi, whose duties in the temple are doubtful, but who sometimes were physicians; the Kleidouchoi, or key-bearers, who perhaps were originally a class of superior door porters, but who appear later to have assumed priestly functions; the Hieromnemes seem to have had purely secular functions, and in common with the Hieres had charge of all receipts and payments; all were under the rule of the Boule of Epidaurus. The Kaniphoroi (or basket-bearers), and the Arrephoroi (or carriers of mysteries or holy things) were priestesses; perhaps in some measure they acted as nurses. There was also a special religious society termed the Asklepiastes.

Turning now from the priests to the suppliants. These we find came from all parts of the Greek world, and from what ancient writers tell us their numbers were great. Where were they housed? Some, of course, dwelt in the abaton, the women probably in one part, and the men in another, but as I have already pointed out, not more than 120 could find beds there at a time; perhaps the invalid was only housed there at first, and when he began to improve was drafted off to a hostel. Assuming that all the buildings which I have suggested to be hostels were such, they could not accommodate more than some four or five hundred patients. Perhaps the usual number attending may have been only some five or six hundred, while at the great festivals many thousands assembled. Whether this large number were lodged in tents or temporary wooden buildings is uncertain.

The patient on arriving probably had an interview with the priest or other official, and arranged about his accommodation with one of the Hieromnemes, or other secular person. He performs certain rites, bathes in the sacred fountain, and offers sacrifices under the direction of the Pyrophoros; the poor man gives his cake, the rich his sheep, or pig, or goat. When night comes he brings his bed clothing into the abaton and reposes on his pallet, putting usually some small gift on the table or altar. The Nakoroi having come round to light the sacred lamps, the priest enters and recites evening prayers to the god, entreating divine help and divine enlightenment for all the sick assembled there; he then collects the gifts which had been deposited on altars and tables; later the Nakoroi enter, put out the lights, enjoin silence, and command everyone to fall asleep and to hope for guiding visions from the god. The abaton was a lofty and airy sleeping chamber, its southern side being an open colonnade. It is singularly like the "shelter balcony" or "*Liegehalle*" now used in treating phthisis. According to the inscriptions the god frequently appeared in person, or in visions, speaking to the sick man or woman concerning their ailments. Whether these visitations were merely hallucinations in individuals whose imagination had been excited, or whether some priest in the dim light enacted the part of Asklepios; whether the patient was put under the influence of opium or some other drug provocative of dreams, or whether, by some acoustic trick, the priests caused the sick to hear spoken