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 sterilized 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 1-25 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