equals within a fraction of a per cent. that of the ω-group of the lysine. It appears that the ω-group of lysine constitutes practically all of the free amino nitrogen of the native proteins determinable with nitrous acid. The albumoses show appreciably more free amino nitrogen, which is to be expected from the fact that the protein molecule is partially broken down in their preparation.

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An experimental study of anti-anaphylaxis.

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If a guinea-pig be given a single injection of a foreign proteid, it becomes, after the lapse of 10 to 14 days, actively sensitized to that proteid, in such wise that the reinjection of the same, in doses far too small to cause any symptoms in a normal animal, produces almost immediate death with convulsions. If, however, such a sensitized pig, after the sensitizing injection, be given a second dose, too small to induce its death, it immediately passes into a condition of anti-anaphylaxis, in which it is refractory to the foreign proteid in question, and may manifest no symptoms even after the injection of doses toxic to normal animals. This refractory stage lasts for weeks or months. In the same way, an animal may be passively sensitized by the introduction into its veins or peritoneum of the serum of another animal, which has been previously immunized or sensitized to a foreign proteid; in this case, too, the injection of a relatively small, *i. e.*, sublethal, dose of the same proteid into the passively sensitized animal produces a condition of anti-anaphylaxis.

By no experimental device hitherto employed has it been possible to alter this condition of anti-anaphylaxis. The theories offered to explain it are numerous. Friedemann, in his general review of anaphylaxis, in 1911, cites three hypotheses, those of Gay and Southard, of Besredka, and of Friedberger, all of which he proves to be untenable, and offers three other possible explana-

tions. He concludes that anti-anaphylaxis presents a "most interesting, novel, and as yet unexplained phenomenon."

The following experiments appear to us to throw much light on the problem. Guinea-pigs have been actively sensitized, by injection, to a foreign proteid; after the lapse of about two weeks, they have been rendered anti-anaphylactic by the intra-peritoneal injection of a sublethal dose of this proteid. (That anti-anaphylaxis had actually been so induced was demonstrated by the injection of relatively enormous doses of the proteid into controls similarly treated, without toxic effect.) The anti-anaphylactic animals have then been *re-sensitized.* This re-sensitization is accomplished by bleeding to death another guinea-pig, sensitized to the identical proteid, and then injecting his serum, in amounts of from 2 to 5½ c.c., into the veins of the anti-anaphylactic animal. After the lapse of less than 24 hours, the re-sensitized pig has been tested by an intravenous injection of the foreign proteid. In each and every case, the pig, upon undergoing this test, has manifested the typical symptoms of anaphylaxis, with immediate death. In other words, such a pig acts exactly like a normal pig, which has been passively sensitized by the introduction of serum from an anaphylactic pig.

If instead of a sensitized pig, one makes use of the serum from a pig highly immunized by means of repeated injections, re-sensitization of the anti-anaphylactic animal is accomplished in the same manner.

If an actively anti-anaphylactic pig be passively re-sensitized with the serum of a rabbit immunized to the same foreign proteid, he too becomes hypersensitive, and is killed by a subsequent dose of this proteid.

The same experiments can be performed in the case of animals rendered anti-anaphylactic after passive sensitization. If, during the refractory period, say two days after the anti-anaphylactizing dose, they are re-sensitized to the same foreign proteid by the reintroduction of an immunized rabbit's serum, they may be killed in the typical manner by the re-injection of the proteid. The only difference between passive anaphylaxis as primarily and as secondarily induced, is that in the latter somewhat larger doses are required to re-sensitize. The cause of this difference is being investigated and will form the subject of a later report.

These experiments in anti-anaphylaxis were originally performed with horse serum. In order to avoid any possible source of error, however, due to the complex character of this material they were repeated with a solution of crystalline egg albumen which had been four times re-crystallized, and the same results were obtained.

It appears from these experiments that anti-anaphylaxis is a condition in which an animal becomes refractory to the toxic effects of a foreign proteid simply through the exhaustion from his blood of those bodies which induce the reaction. The proof hereof lies in the fact that the simple re-introduction of these bodies with the blood of another sensitized animal restores him at once to his original condition of anaphylaxis. The bodies which induce the reaction are, so far as we know, two: first, the "anaphylactic anti-bodies," which resemble in character, and may be identical with, the amboceptors; second, the complement substances of the blood.

It is unlikely that the complement is the substance herein at fault, inasmuch as Friedemann states, as the complement is rapidly restored to its normal amount in anti-anaphylactic pigs, whereas the condition of anti-anaphylaxis persists. We have tested whether the complement plays an important rôle by two sets of experiments. In the first place, the animals have been simultaneously sensitized by hypodermic injection to two different forms of foreign proteid, namely horse serum and egg albumen. They have then been rendered anti-anaphylactic to one of these. Immediately, thereafter, it has been found that their sensitiveness to the other albumen is little, if at all, impaired. This indicates that the complement cannot be deficient. In the second type of experiments, the pigs, having been made anti-anaphylactic to egg albumen, have been given an intra-venous injection of 5 c.c. of normal guinea-pig serum, which would, of course, suffice to supply any defect of complement. When now re-injected with egg albumen, they fail to evidence any sensitiveness. This indicates, therefore, that something other than complement, necessary to the anaphylactic reaction, has been removed from the blood in anti-anaphylaxis. It seems to us, therefore, that animals are anti-anaphylactic simply through the absence of the appropriate

anti-bodies from the serum. If these are supplied, an animal passes directly into the anaphylactic condition again, and this reversal could conceivably be repeatedly renewed.

CONCLUSIONS.

1. Guinea-pigs which have been rendered actively anaphylactic by a preliminary injection of foreign proteid (horse serum, egg albumen), and have then been made anti-anaphylactic by the injection of a sub-lethal dose of this proteid, may be immediately re-sensitized by the introduction of serum from a pig sensitized, or one immunized to the same proteid. Death is then produced, with typical anaphylactic symptoms by the intravenous injection of an amount of this proteid which to normal pigs is non-toxic.

2. Actively anti-anaphylactic pigs may be re-sentitized with serum derived from a rabbit immunized against the same proteid.

3. Passively sensitized pigs, rendered anti-anaphylactic by the usual methods, can be re-sensitized by the re-introduction of an immune serum.

4. A pig simultaneously sensitized to two different foreign proteids, may be rendered anti-anaphylactic to one of these by the usual methods, while its sensitiveness to the other remains unimpaired. This demonstrates that anti-anaphylaxis cannot be due to exhaustion of complement, since complement is amply present.

5. The introduction of normal guinea-pig's serum into an anti-anaphylactic animal fails to re-sensitize. Some factor other than complement, therefore, must be introduced in re-sensitization.

6. The absence of available anaphylactic antibodies appears to be the cause of the refractory condition known as anti-anaphylaxis, and re-sensitization is due to their re-introduction.

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The toxicity of foreign leucocytes.

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The injection of from 0.7 c.c. to 1.0 c.c. of rabbit leucocytes into the cerebral meninges of dogs is apparently invariably fatal.