From Tables II and III it will be seen again that the normal-fed group showed a significant fall in the sugar level after the second hour. This fall corresponds to the decrease obtained at the time of recovery from the anesthesia, the duration of which averaged 2½ hours after the injection. The starved group again shows a smaller fall, but since the difference between the initial and final values is almost 2 times the deviation of the difference, the change is probably significant. The time at which this fall occurs also corresponds to the time of recovery which, in the starved group, occurred from 3 to 4 hours after the injection.

The drop in the blood sugar of normal-fed rabbits at the time of recovery from nembutal anesthesia brought the blood sugar to the initial level of the animals which were fasted 24 hours. But the normal-fed animals were recovered at the time of the drop in the blood sugar, while the fasted rabbits with the same sugar level remained anesthetized. There is, therefore, no evidence of a correlation between the blood sugar level, *per se,* and the susceptibility to nembutal. It appears that inanition has some effect other than that of lowering the blood sugar. Some of the metabolic processes are altered so as to render the animal susceptible to the drug for a much longer period of time. Since the starved group as well as the normal-fed show a fall in the sugar level at the time of recovery, it appears that the nembutal has some effect upon carbohydrate mobilization. We are at present studying the possible relationship between liver function and this increased susceptibility to nembutal anesthesia.

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A Contribution to Drug Allergy: Antipyrine.

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Landsteiner and Lampl[1] showed that new protein antigens could be formed through the chemical union between chemically simple drugs, such as anilin, and a protein. This is true for precipitin reactions in rabbits, as well as for shock experiments with guinea pigs (Landsteiner[2]). The sensitized animals do not react to an

[1] Landsteiner and Lampl, *Z. Immun. Forsch.*, 1917, **21**, 193.

[2] Landsteiner, *J. Exp. Med.*, 1924, **39**, 621.

injection of the simple compounds uncombined to protein. On the other hand, they may react to a protein other than the one used for the sensitizing injections, provided this new protein has linked to it the same chemical compound which had been added to the new antigen. It remained to be determined whether the results obtained by precipitin reactions and anaphylactic shock apply to isolated organs as well. For this purpose, experiments were performed with the isolated guinea pig uterus, according to the method of Schultz and Dale.

Antipyrine was diazotized and coupled to protein, after the method of Landsteiner and Lampl. Two antigens were made. In one, the diazo-antipyrine was combined with rabbit serum proteins, and used for injections into rabbits. In another, the diazo-antipyrine was coupled to guinea pig serum proteins, and used for injections into guinea pigs. In this way the interfering presence of antigenic protein was eliminated.

Positive precipitin reactions were obtained against rabbit serum which had been treated with antipyrinediazorabbit protein. Precipitation occurred when the sera were tested to diazoantipyrine-protein antigens of rabbit, guinea pig or egg white proteins. Pure antipyrine added to the precipitin tubes inhibited the precipitin reaction from these antigens.

The uteri of virgin guinea pigs which had been "immunized" to antipyrine-diazo-guinea pig protein were suspended *in vitro,* after the technic of Dale.[3] Wherever it was possible, each uterus was cut into 4 sections, and each segment suspended individually. Each uterus could be subjected to the following procedures:

(a) If no spasm followed the instillation of the antigen, a larger dose was added to the second strip, and more to the third and fourth, until either spasm occurred or the strip designated as insensitive.

(b) If spasm did follow the instillation of the antigen, then the second strip had added to its 100 cc. of bath fluid, 10 mg. of antipyrine, the haptene group.

(c) A third strip of a sensitive uterus was treated with an antigen formed of diazo-antipyrine and a protein other than the one used for the induction of sensitivity.

It was routine to treat a sensitive strip of the uterus, after spasm had been induced, and the Ringer's solution replaced, with a second dose of antigen. The specificity of the first spasm is proven by the lack of effect of the second dose. At the end of each experiment, barium chloride or pilocarpine was instilled into the bath, in order

[3] Dale, *J. Pharm. Exp. Therap.*, 1912, **4**, 167.

to test the ability of the uterine strip to respond to an adequate stimulus.

Twenty guinea pigs were injected with antipyrineazoguineapig serum protein antigen. Of these, 15, or 75%, proved sensitive by the uterus method described above. Of the 15 sensitive uteri, 14 were rendered insensitive to the homologous antigen by the previous treatment with 10 mg. of antipyrine added to the 100 cc. of bath fluid. That the antipyrine has a specifically depressant action was shown by the following experiment. Five guinea pigs were injected with eggwhite solution. The uteri were shown to be sensitive in 4 of these. The specific spasm which followed the antigen was uninfluenced by the previous addition of 10 mg. of antipyrine to the bath. *Therefore the previous inhibition of the tissue response by the antipyrine was specific.* This is in conformity with the inhibition of the precipitin reactions by the antipyrine.

None of the strips found sensitive reacted to a second dose of the antigen, proving the specificity of the reaction. The haptene desensitizes *without* previous spasm, while the antigen does so after specific spasm. Desensitization of this tissue cannot therefore be accounted for on a theory of histamine depletion resulting from the spasm.

All of the sensitive guinea-pig uteri responded by spasm also to antipyrine-azo-rabbit serum, or antipyrine-azo-eggwhite protein. In these cases only the azo-antipyrine group had any relationship to the original antigen. These findings are parallel with those obtained with the precipitin reactions in rabbits.

The uteri of 2 of these animals were removed surgically, the animal being allowed to recover. The uteri were sensitive to our antigen. Two days later the surviving pigs were injected intravenously with the antigen. Both animals showed signs of anaphylactic shock. One died within 30 minutes, in typical anaphylactic bronchospasm, as revealed at autopsy. The other survived. Protection experiments in intact pigs was not tried, because the antipyrine proved too toxic to administer in sufficient doses, and for lack of material.