from these types may occur, depending on the growth characteristics of the plant, the severity of the injury, and the stage to which the plant has developed before it is attacked. Swellings or galls, comparable to those caused by the root-knot nematodes *Meloidogyne* spp.,[1] do not occur.

The aboveground symptoms are essentially the same as those caused by any condition that deprives a plant of an adequate root system. Growth is retarded, the foliage wilts easily, and the plant has little ability to withstand drought. The foliage of severely stunted plants frequently turns yellow, a condition that may be quite pronounced on corn. It seems probable that such a chlorotic condition is not caused directly by the nematode but is due to some soil deficiency or other condition that is aggravated in the plant through lack of a normally functioning root system.

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[1] Formerly *Heterodera marioni* (Cornu) Goodey.

Attempt to Show Diffusion of Essential Growth Factors from an Induced Penicillin-resistant Culture to the Parent Penicillin-sensitive Strain

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In 1940 Woods (*1*) showed that PABA, an essential growth factor for many bacteria, was competitively replaced by sulfanilamide, thus inhibiting the susceptible organism. He also demonstrated that bacteria capable of synthesizing PABA were resistant to sulfanilamide, indicating the production of a diffusible metabolic substance to inhibit a chemotherapeutic agent.

Bailey and Cavalitto (*2*) stated that penicillin acts by blocking essential sulfhydryl and possibly amino groups, producing a metabolic block.

Gale and Rodwell (*3*) believe that penicillin-sensitive organisms cannot synthesize many amino acids, especially glutamic acid, whereas resistant organisms can synthesize them from inorganic constituents; penicillin thus acts to impair the ability to assimilate glutamic acid from the medium.

Plough and Grimm (*4*) converted a naturally penicillin-resistant heterotrophic strain of *Salmonella typhimurium* to a penicillin-sensitive cysteine-requiring mutant, indicating that penicillin blocks assimilation of certain amino acids required by the susceptible strain.

Hunter and Baker (*5*) objected to the assimilation-blocking theory of Gale by revealing that a penicillin-sensitive *Bacillus subtilis* may synthesize and not assimilate amino acids, and that some strains of *Escherichia coli,* which synthesize but do not assimilate amino acids, are inhibited by penicillin in high concentration.

In the present study an attempt was made to determine whether an induced penicillin-resistant strain of *Staphylococcus aureus* could synthesize a diffusible product that might supply the parent (penicillin-sensitive) strain with the essential nutrilites to grow in the presence of a lethal amount of penicillin.

The culture used was *Staph. aureus* strain L obtained from Boston University School of Medicine and did not produce penicillinase. The penicillin used was sodium penicillin G (Commercial Solvents Corporation). Resistance was induced as previously described (*6*) by transferring 0.5 ml from the tube of the lowest concentration of penicillin that inhibited to 10 ml of plain broth. After 24 hr of growth the broth culture was used in a penicillin titration of a higher range.

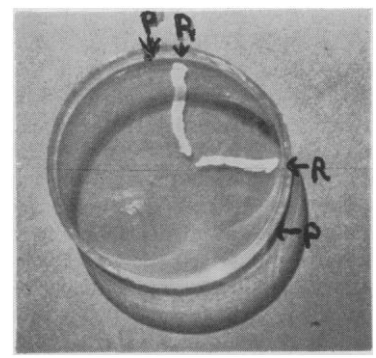

FIG. 1. Penicillin-agar plate. *P,* culture sensitive to penicillin; *R,* culture resistant to penicillin.

To determine possible diffusion of essential nutrilites from the resistant to the sensitive strain, a method described by Davis (*7*), which he used to show diffusion of synthesized products from one culture to a biochemically deficient mutant, was employed. A typical plate is shown in Fig. 1. Nine ml of plain agar at 45°–50° C was added to 1.0 ml of the appropriate concentration of penicillin in a Petri dish. The agar and penicillin were mixed by rotating, and the agar was allowed to harden. Streaks of 24-hr broth cultures of resistant and parent strains were made adjacent to each other to test for diffusion of vital materials from the resistant culture to the sensitive culture. Controls of parent and resistant strains were prepared on the same plate by making streaks at distant parts of the plate.

It appears that generally no essential nutrient was supplied by diffusion to enable the penicillin-sensitive strain to reproduce adjacent to the penicillin-resistant strain. These results favor the theory of Gale and Rodwell (*3*) that penicillin impairs the ability of the culture to assimilate an essential nutrient from the medium, whereas the resistant strain is able to synthesize the substance within its cell. It must also be considered, however, that other mechanisms may be taking place, such as the "learned" ability of the resistant

mutant to grow in the absence of an essential factor without producing this factor.

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An Antidiuretic Substance Obtained by Digestion of Globulin with Pepsin[1]

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Croxatto and Croxatto (*1*) obtained a hypertensive principle (pepsitensin) by treating with pepsin the globulin fraction of plasma that contains hypertensinogen. Also, under these conditions, another substance was formed, with a marked oxytocic effect (*2*). A third substance, which produces an intense antidiuretic action, can be obtained if the extracts containing pepsitensin are digested again with pepsin. This last substance was detected and titrated, using a method described by Burn (*3*).

Rats were used under a water load (5% of body weight), and the different extracts were injected intraperitoneally. Each dose was injected in 4 animals, and the volume of urine measured every 15 min during a period ranging from 2 to 3 hr. Diverse fractions of plasma obtained by precipitation of mammalian blood (human, horse, and ox blood) with $(NH_4)_2SO_4$ were used. These fractions were dialyzed, acidified to pH 3.5, and treated with pepsin (10 mg/100 ml) for 4 hr at 37° C. The fractions were added to 2 volumes of boiling alcohol. The precipitate was then separated, the solvent evaporated, and the final aqueous extracts were reduced to a third of the original volume.

Typical pressor action of pepsitensin was observed in cats and rats, especially after the injection of extracts prepared from fractions that contained hypertensinogen. These extracts also caused an intense oxytocic effect on the isolated uterus of guinea pigs. No antidiuretic effects, or very slight ones, were observed if the extracts were injected intraperitoneally in the rat. An intense antidiuretic action was observed after the injection of the extracts if they were submitted to a second digestion with pepsin (pH 2.5–3.5) for 24 hr, filtrated, and neutralized. It is interesting to notice that these extracts lost their pressor action, maintaining or increasing the oxytocic effect (Figs. 1, 2).

It is possible to assume that the antidiuretic action may be caused by a peptide. It dialyzes slowly through cellophane membranes and is unstable at an alkaline pH.

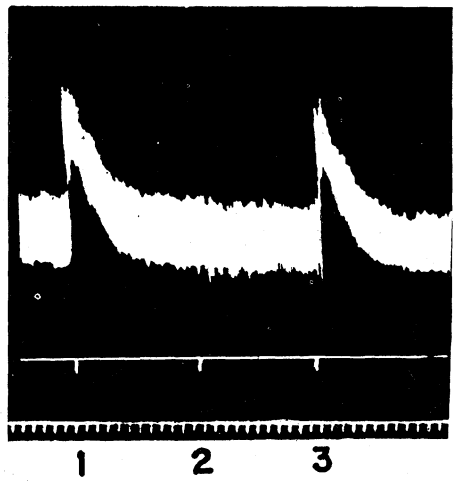

FIG. 1. Blood pressure recording: (1) 1 ml of extract obtained from 7 ml of horse hypertensinogen, plus 0.8 mg of pepsin incubated 4 hr at 37° C; (2) 1 ml of the same extract, following injection after a second digestion with 3 mg pepsin; (3) same as (1).

In spite of the fact that it was not established whether some antidiuretic substance preexists linked to globulins, we believe that most of it is originated by enzymatic hydrolysis from a globulin precursor, as it occurs with hypertensin. If hypertensinogen is treated with HCl (adding no pepsin), no antidiuretic substance is formed. Small yields of antidiuretic substance were obtained from either albumin or fibrinogen.

Our findings indicate that some of the antidiuretic substance obtained from body fluids may have an extrapituitary origin, as has been pointed out by Walker (*4*). Furthermore, these results would also suggest a close relationship among pepsitensin, hypertensin, vasopressin, and oxytocin. Other studies have shown important links between the mechanisms that inactivate neurohypophysis hormones and hypertensin (*5*). Such observations are of interest, especially after the results

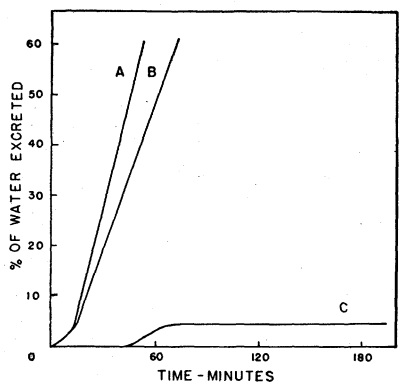

FIG. 2. Urinary excretion curve obtained from 3 groups of rats under a water load (5% body wt). *A*, control group; each rat was injected at 0 with 1 ml of 0.9% NaCl. *B*, group that received 1 ml of the extract injected at 1, Fig. 1. *C*, corresponds to the group that received the extract injected at 2, Fig. 1.

[1] A complete report of this work will be published in *Acta Physiologica Latinoamericana.*

[2] We are grateful to the Fundación Gildemeister for financial help.