

of carbon-14 in serine and alanine isolated from the silk fibroin, can help us to decide which of these routes is the most important in *Bombyx mori*. We already know that the administration of glycine-1-¹⁴C is not followed by a redistribution of the labelled carbon between the two carbons (see preceding communication).

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¹ Dunn, M. S., Camien, M. N., Rockland, L. B., Shankman, S., and Goldberg, S. C., *J. Biol. Chem.*, **155**, 591 (1944).

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Cellulolytic Activity of Phytopathogenic Bacteria

As the great majority of phytopathogenic bacteria are extracellular parasites occupying chiefly the vessels and the intercellular spaces, and causing vascular diseases, soft-rots, spot diseases, etc., the ability to decompose the surrounding walls composed of pectin and cellulose is supposed to be an important factor controlling the destruction and/or enlargement of the tissues attacked. Although Husain and Kelman¹ reported the presence of cellulase in diseased tomato tissues infected by *Pseudomonas solanacearum*, our knowledge of the cellulolytic enzymes is poor in comparison with that in the field of pectolytic enzyme studies of plant pathogens.

We have sought evidence *in vitro* as to whether cellulolytic enzymes are produced by other phytopathogenic bacteria. Bacterial species or isolates used were from our collections in Japan. For the recognition of cellulolytic enzymes carboxymethylcellulose gel medium was used as stab cultures and observed periodically for three weeks at an incubation temperature of 30° C. When the enzymes were produced, stratiform liquefaction of the gel followed the growth of bacterial species inoculated. The carboxymethylcellulose gel medium was prepared as follows: 2 per cent of carboxymethylcellulose (Koso Chem. Co., Ltd., Tokyo) was dissolved in hot basal medium (70° C.) such as bouillon, potato decoction, synthetic solution A (ammonium dihydrogen phosphate, 1 gm.; potassium chloride, 0.2 gm.; magnesium sulphate, 0.2 gm.; distilled water, 1 l.); synthetic solution B (ammonium nitrate, 1 gm.; potassium hydrogen phosphate, 1 gm.; magnesium sulphate, 0.2 gm.; distilled water, 1 l.); or synthetic solution C (Ushinsky's solution without glycerol), then heated almost to boiling point and stirred sufficiently to form a homologous gel. The mixtures were placed in tubes and autoclaved at 15 lb./in.² for 10 min. Tests were made with 13 species of *Pseudomonas*, 7 species of *Xanthomonas*, 2 species of *Erwinia*, 1 species of *Agrobacterium* and *Corynebacterium*, including various isolates of each species.

Prompt liquefaction of the gel was observed in bouillon and potato decoction cultures after 3-5 days, while in synthetic media the gel was liquefied very slowly and some isolates showed only partial liquefaction even after three weeks. The liquefying species or isolates grew well throughout the dissolved layers of gel. Growth of the non-liquefying ones was moderate on the gel surface in bouillon and potato decoction, but feeble or absent in synthetic media.

The results, obtained from potato-decoction gel cultures, showed that cellulolytic enzymes are produced from *Ps. panici* (syn. *Ps. setariae*), *Ps. solanacearum*, *Xanthomonas campestris*, *X. citri*, *X. nigromaculans*, *X. oryzae*, *X. pisi* (a new species, not reported), *X. pruni*, *X. vesicatoria*, *Erwinia carotovora* (including *E. aroideae* as a synonym), *E. milletiae*, *Corynebacterium sepeдонicum*, but not from *Ps. aeruginosa*, *Ps. andropogoni*, *Ps. cichorii*, *Ps. eriobotryae*, *Ps. iridicola*, *Ps. maculicola*, *Ps. marginalis*, *Ps. marginata*, *Ps. phaseolicola*, *Ps. syringae*, *Ps. tabaci*, *Ps. theae* and *Agrobacterium tumefaciens*.

In the case of *X. oryzae*, *E. carotovora* and *Ps. solanacearum*, the rapidity of liquefaction varied with the kinds of isolates used.

The pathological significance and biochemical characteristics of the cellulolytic enzyme produced by these bacteria will be discussed elsewhere.

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The Kidney and Erythropoiesis

It has been pointed out^{1,2} that, in the dog, bilateral nephrectomy abolishes erythropoiesis, and it was suggested that the kidney produces a factor stimulating red blood cell formation. The experimental result did not definitely exclude retention of some toxic substance that inhibits the erythropoiesis, since the nephrectomized animals were still uræmic (blood urea level at about 300 mgm. per cent) despite peritoneal dialysis.

In further experiments an attempt was made to dissociate a toxic effect from the lack of some erythropoietic factor of renal origin. Thus, erythropoiesis was studied in a group of eight dogs submitted to bilateral ureter ligation. This group was compared with one group of seven intact and one group of eight bilaterally nephrectomized animals.

Bone-marrow samples were examined every other day before and after surgery. 72 hr. after surgery, 5 µc. of iron-59, previously incubated in 20 ml. plasma, were given intravenously. The uptake of iron-59 into erythrocytes was measured in eight dogs during the three following days.

Five dogs lived long enough to allow studies of the maximal incorporation of iron-59. The dogs with ligated ureters as well as the nephrectomized ones refused food, and were given two or three daily peritoneal dialyses, thus reaching similar levels of toxic retention and malnutrition.

Despite these conditions, the following experiments show that erythropoiesis is almost normal in animals with ureteral ligation, showing that the absence of a kidney-factor is responsible for the impaired erythropoiesis in nephrectomized dogs.

The disappearance curves of iron-59 from the plasma are similar in normal dogs (39-153 min.) and in those with ligated ureters (64-171 min.). The average time at which half the radioactivity initially present has disappeared (*T*/2) is 299 min. in the nephrectomized, 89 in the normal animals and 96 in those with ligated ureters.

The rate of turnover of plasma iron is higher in animals with ligated ureters (0.25-0.85 mgm./kgm./day; mean, 0.48) than in nephrectomized ones