generalize results from one laboratory to another, our results indicated the need for a note of caution. This is particularly true if one also considers that any one of these cultures may show several forms of sensitivities to X-rays and ultra-violet light, depending on the growth medium and other factors⁵. The cultures used in this work were not pre-selected for heterogeneity. They were chosen because they were readily available. It is likely that other groups of *B/r* cultures from other laboratories would be equally heterogeneous. Greater attention to the techniques of maintaining cultures and periodic checking of culture characteristics may allow a more meaningful correlation of the results obtained in different laboratories. We suggest that the designation *E. coli B/r* be reserved only for cultures having all the characteristics originally observed by Witkin.

HOWARD I. ADLER
SARA D. HASKINS

Biology Division,
Oak Ridge National Laboratory,
Oak Ridge, Tennessee.

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These bacteria were subjected to detailed examination. In addition to them the capacity of breaking down the chitin of rhizosphere bacteria isolated on non-chitinous media by other workers was also studied: none of the five *Pseudomonas* strains belonging to various serological groups, grown from the rhizosphere of flax by Lovrekovich⁵, tested with the method described above, was found to decompose chitin. Out of the sixteen *Bacterium* strains grown from sugar-beet, Gyurkó⁶ subjected to detailed physiological examination five strains proved to be chitinivorous and the strains kept on meat agar have preserved this property for more than two years.

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KECSKÉS MIHÁLY
Research Laboratory for Soil Biology,
Hungarian Academy of Sciences,
Sopron.

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VIROLOGY

Influenza Virus Sialidase

THE sialidase of influenza virus has been investigated quantitatively using the method recently described by Warren¹ for determining free sialic acid in the presence of the compound bound to mucoprotein substrates. For this work, the substrate used was a mucoid derived from the nest-cementing substance of the swiftlet (*Collocalia* sp.). The crude material, available as Chinese 'edible birds' nest', was extracted with water at 60–65° to yield a product which contained 10 per cent acid-hydrolysable sialic acid and which in high titre inhibited the agglutination of chicken erythrocytes by 'indicator' virus. Active virus was sedimented from freshly gathered chorioallantoic fluid by centrifugation at 35,000g for 90 min. The virus pellet was re-suspended in water to a volume 1/50–1/100 that of the original fluid, effecting thereby a corresponding increment in haemagglutinin titre as well as separation of virus from interfering chromogens present in chorioallantoic fluid.

Aliquots of virus (A2/Japan/305/57) representing a six-fold concentration of infective chorioallantoic fluid were mixed with substrate in phosphate (*M*/15) buffers, pH 4.0–7.8, at 37°, in the proportion 10 µgm. viral nitrogen to 1.4 mgm. substrate. Samples were removed at 10-min. intervals from 0 to 50 min., and were immediately heated to 75° for 10 min., a temperature found to inactivate the virus completely. All samples were either stored frozen or at 4° prior to being analysed simultaneously for sialic acid. Virus alone and solutions of substrate at each pH, after incubation and heating to 75° for 10 min., showed no free sialic acid. The most rapid evolution of chromophore due to enzyme occurred at pH 5.3 and 6.1. These values probably encompassed the pH optimum for this strain, as the enzymatic release of sialic acid at pH 4.0, 7.0 and 7.8 was significantly slower (Fig. 1A). Closely similar results were obtained with three other strains of virus (NWS, FM1 and PR301) and are therefore not given in

Chitin Breakdown of Rhizosphere Bacteria

BENECKE¹ was the first to isolate chitinivorous bacteria from Kiel harbour. Since then much work has been done on microbiological breakdown of chitin. Micro-organisms producing chitinase have been observed in fresh and salt waters, in various kinds of soils, in plants and animal bodies, both living and decomposing. It has been reported that pathogenic micro-organisms also break down chitin.

I have studied whether bacteria living in the rhizosphere of superior plants are capable of attacking chitin.

For this purpose chitin has been prepared from the wing-cases of *Melolontha melolontha* L. with the Zechmeister procedure applied also by Gehring². The native chitin obtained in this way was suspended in medium according to Bucherer³. The chitin was present in the medium as the sole source of carbon and nitrogen in amounts of 0.1, 0.25, and 0.5 per cent. Dipotassium hydrogen phosphate and magnesium sulphate (0.03 or 0.1 per cent) were added. The solid medium contained 2 per cent washed agar. In all cases the pH was 7.

Bacteria were isolated on chitin agar and meat agar plates from the rhizospheres of winter wheat (*Triticum vulgare*), sugar-beet (*Beta vulgaris saccharifera*), and false acacia (*Robinia pseudoacacia*) with Tepper's⁴ successive washing method (from the dilution of washed and triturated root). About 20 per cent of the bacterial colonies grown on meat agar, tested on both solid and liquid media, proved to be chitinivorous.

The bacterial colonies grown on chitin agar did not show a great variety of types, but the number of individuals was rather high. Their total reached several hundred thousands per 1 gm. of washed root.

From the chitinivorous rhizosphere bacteria of winter wheat and false acacia a rod-shaped, most frequently occurring, active chitinivorous organism—one from each—was selected. These retained their capacity of breaking down chitin for rather a long time.