

Catches made over a period of years indicated that *S. neavei* adults seem to prefer to live near the smaller rivers and streams, and accordingly work was begun on a small river passing through heavily wooded country, and having many small cascades and waterfalls in its course. Large numbers of the younger stages of various insects, as well as crabs and fish, were examined during a period of two weeks for evidence of such an association, with negative results. However, searches in two of the larger rivers, the Kipsonoi and the Sondu, soon produced evidence that there is a definite association between freshwater crabs (*Potamonautes (Potamon) sp.*) and a *Simulium* sp. As many as ten larvæ have been found attached to the carapace of one crab, and the maximum number of pupæ observed was three. Adults bred from these pupæ appear to be *S. neavei*, and the male and pupa will be described in detail at a later date; the pupal respiratory organ has eight long slender breathing filaments, equal in length to the cocoon. The female has already been described by Roubaud.

The association between crabs and this *Simulium* species coincides with the distribution of *S. neavei* adults, whereas that between *Afronurus* and *Simulium* species seems to occur at a higher altitude well outside the *S. neavei* belts. Only one specimen of *Afronurus* collected in the Kipsonoi was found to have a pupa attached, and not more than twelve were collected in the Sondu during a period of six days; but in the higher Kiptiket River, 20 miles away and some 300 ft. higher, twenty nymphs with pupæ attached were collected during a period of two hours.

The crabs concerned seem to prefer to live in the rockier parts of the rivers, such as cascades, and no specimens have yet been caught on mud or other soft river-bed material.

Simulium-Afronurus associations have now been found in several Kenya rivers, and the whole problem is still the subject of study in all its aspects. Dr. G. Marlier, of I.R.S.A.C., Belgian Congo, has informed one of us (V. D. v. S.) *in litt.* that he has found similar *Simulium-Afronurus* associations in rivers in the Kivu Province of the Congo, apparently two species of *Simulium* being involved; Dr. Marlier is publishing a note on this subject elsewhere.

In conclusion, we wish to thank Mr. Hugh Copley, fishwarden, Kenya Colony, and Dr. T. Farnworth Anderson, director of Medical Services, Kenya Colony, respectively, for permission to publish this joint note.

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Sulphamic Acid as a Test for Nitrate-reducing Bacteria

SULPHAMIC acid (aminosulphonic acid) decomposes nitrites quantitatively¹ with evolution of nitrogen gas according to the equation



The reaction, which is very specific and strongly catalysed by hydrogen ions, may be used for the removal of interfering nitrites, for example, when testing for nitrates², or in water analysis as part of the Winkler method for dissolved oxygen determina-

tions, where nitrites will cause serious error unless removed. For this latter purpose, according to Cohen and Ruchhoff³, sulphamic acid possesses distinct advantages over sodium azide.

It seemed worth while investigating whether sulphamic acid had any value in bacteriology as a reagent for detecting nitrites in the nitrate-reduction test, for which it would appear it has not hitherto been used. Preliminary experiments to ascertain the limits of sensitivity of the reagent were carried out using a solution of sulphamic acid prepared according to the directions of Cohen and Ruchhoff (*loc. cit.*). This is a 4 per cent solution in 20 per cent v/v sulphuric acid. Standard solutions of nitrite were prepared from silver nitrite. The following technique was used. 1 ml. of standard nitrite was pipetted into a small test tube (7.5 cm. long by 8 mm. internal diameter). The tube was slightly inclined and 1 ml. of the sulphamic acid solution was pipetted down the side. By this method water blanks showed no bubble formation. In the test solutions gas bubbles were readily seen in concentrations of nitrite down to 5 p.p.m. (expressed as nitrogen).

One hundred cultures of catalase-positive cocci were tested for nitrate reduction both by the sulphamic acid technique outlined above and also by the usual modification employing the Griess-Ilosva reagents (sulphanilic acid and α -naphthylamine in acetic acid solution). All the cultures were examined by one test before the second test was applied. In this way, the result of one test on an individual culture was not known before applying the second test, thus avoiding any prejudgment of the result. The records showed that in each of the sixty-two cultures in which a positive result was noted with the Griess-Ilosva reagents, evolution of gas was observed with the sulphamic acid solution. No gas bubbles were seen in the remaining cultures.

A full account of the work and a critical survey of the two methods will be published elsewhere.

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¹ Yost, D. M., and Russel, jun., H., "Systematic Inorganic Chemistry" (Oxford University Press, 1946).

² Feigl, F., "Qualitative Analyses by Spot Tests" (3rd (English) edit., Elsevier Pub. Co., 1947).

³ Cohen, S., and Ruchhoff, C. C., *Indust. and Eng. Chem., Anal. Edit.*, **13**, 622 (1941).

Effect of Aspartic Acid on Growth of Plant-Virus Tumour Tissue

RECENTLY several workers have concluded that, under certain conditions, amino-acids can be assimilated intact by yeasts^{1,2}. White and Munns¹ conclude that aspartic acid can be used as a source of nitrogen by yeast, and also that the carbon skeleton can be used if fermentable sugars are present (probably as energy sources). They found that when aspartic acid is added to the test medium in sufficient amount, the growth of yeast greatly exceeds that when only hexose sugars are present.

Results of this nature have been found by Nickell and Burkholder³ working with virus tumour tissue from the sorrel plant (*Rumex acetosa* L.)^{4,5} grown *in vitro*. It was found that aspartic acid could serve, to a certain extent, as a carbon source for the growth of this tissue. Nitrate was found to be the best source of nitrogen; but aspartic acid,