binary division of the protoplasm, (3) in the possession of stages in multiple cleavage not described by Davis for *D. salmonis*, and (4) by the difference in host, *D. salmonis* being parasitic on gills of the chinook salmon.

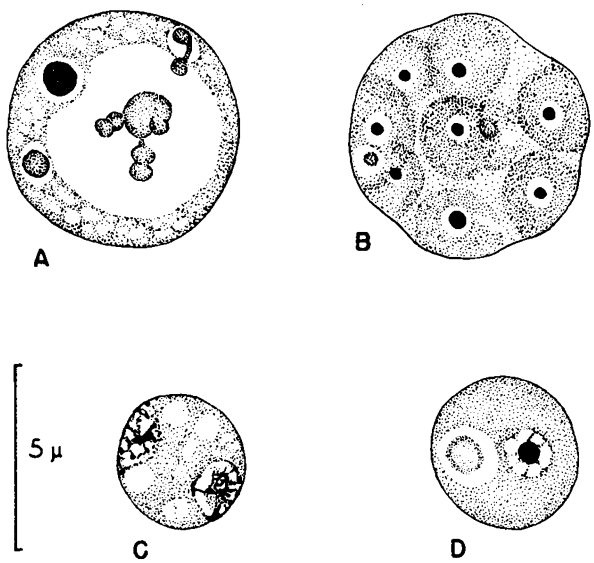

FIG. 1. Drawings from specimens stained with Heidenhain's iron haematoxylin and eosin. A—A mature spore with markedly irregular vacuoplast, cytoplasmic inclusions, and very large vacuole. B—Multiple fission resulting in several daughter cells. C—A binucleate stage with chromatin in diffuse condition, and showing beginning vacuolation of the cytoplasm. D—An immature spore with small vacuole and vesicular nucleus. The radiating fibers from the endosome are not commonly seen.

All other described species of *Dermocystidium* have hosts confined to fresh-water environments, the estuarine environment of *D. marinum* being unique.

A small body of literature has been built up on *Dermocystidium*. At the present time the known species include *D. pusula* (Pérez) 1907, *D. branchialis* Léger (*7*), 1914, *D. ranae* Guyénot and Naville (*4*), 1921–22, *D. daphniae* Jirovec (*5*), 1939, *D. vejdovskyi* Jirovec (*6*), 1939, and *D. salmonis* Davis (*3*), 1947. Hosts include various species of salamanders of the genus *Triturus*, frogs of the genus *Rana*, several fishes including trout, pike, and salmon, and one invertebrate, *Daphnia magna*. All species excepting one have been reported from localities in France, Germany, and Ireland. One species only is recorded in the United States, *D. salmonis* Davis, from the Sacramento River, California.

Several closely related genera are known, but the evaluation of most of the relationships is a problem for the future. At present it seems certain that *Blastocystis* Alexeieff 1911 is the nearest well-established genus. The relationship is based on certain parallels of staining reaction of the nucleus and the vacuoplast. For an exhaustive treatment of staining reactions of *Blastocystis* see Bach and Kiefer (*2*), 1923, and for *Dermocystidium* see Perez (*10*), 1913. Also, there are definite parallels of developmental cycle including production of what Alexeieff (*1*) considered to be ascospores.

Data acquired thus far independently by each author show the association of this organism with dead or dying oysters under certain environmental conditions, the limits of which can be reasonably well defined. The chief controlling factors appear to be temperature and salinity, low temperature and low salinity evidently retarding the development of the infestation.

Oysters in all conditions of health are susceptible to infestation, but factors resulting in fatigue, such as spawning, and miscellaneous adverse environmental conditions accelerate the intensity and spread of the organism.

Investigations of the taxonomy, physiology, ecology, and distribution of this parasite covering the geographical range of *Crassostrea virginica* are still in progress. A full report will be published elsewhere at a later date.

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Action of Penicillin on Bacterial Utilization of Amino Acids and Peptides[1]

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In a previous communication (*5*), there was reported the isolation of a rodlike, Gram-negative bacillus from a contaminated solution of L-leucylglycine in 0.9% NaCl. This organism, termed strain SF, grew satisfactorily in a solution of 0.1 M L-leucylglycine in physiological saline, but did not grow when the dipeptide was replaced by a mixture of 0.1 M L-leucine and 0.1 M glycine. Only poor growth was obtained in the presence of 0.1 M L-leucine. Addition of glucose to these media did not alter appreciably the extent of bacterial growth.

The growth response of strain SF has now been studied in a more complex basal medium which contains, per liter, 5 g NaCl, 0.2 g $MgSO_4 \cdot 7H_2O$, 2 g KH_2PO_4, 2 g K_2HPO_4, and 1 ml of the trace element solution described in reference *3*. Maximal growth in the presence of L-leucylglycine, dissolved in this medium, was attained at a dipeptide concentration of 0.005 M, and subsequent growth studies with the dipeptide or its component amino acids were conducted at this molar concentration for each test compound. In these experiments, Evelyn colorimeter tubes containing the test solutions (final volume, 10 cc) were inoculated with approximately 10^8 organisms, and the slanted tubes were shaken (120 oscillations per min) at 30° C. The extent of bacterial growth was measured

[1] This study was aided by a grant from the Rockefeller Foundation.

turbidimetrically in an Evelyn colorimeter (filter No. 540).

In contrast to the failure of strain SF to grow on a mixture of L-leucine and glycine in 0.9% NaCl, bacterial growth is observed when these amino acids are present in equimolar proportions (0.005 M) in the new medium. Whereas the final turbidity ultimately attained under these conditions is equal to that observed in the presence of an equivalent quantity of L-leucylglycine, growth on the amino acid mixture is considerably slower than with

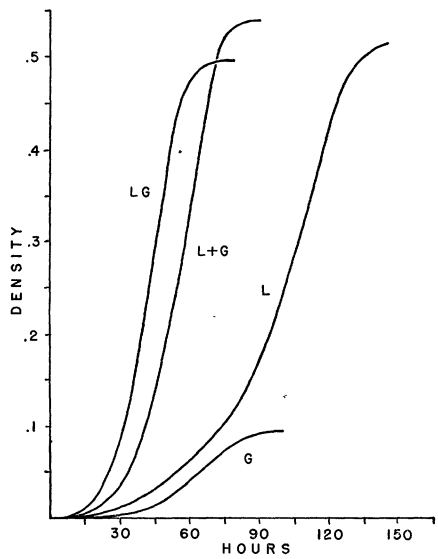

FIG. 1. Representative growth curves for strain SF in the presence of L-leucylglycine (LG), of a mixture of L-leucine and glycine (L + G), of L-leucine (L), and of glycine (G). The concentration of each test compound was 0.005 M. To facilitate inspection of the curves, the observed density readings (2-log galvanometer reading) have been omitted.

the dipeptide (cf. Fig. 1). As will be seen from the data in Fig. 1, strain SF also can grow, albeit slowly, either on 0.005 M L-leucine or on 0.005 M glycine. The comparatively small extent of bacterial growth observed with the latter amino acid may be attributed to the fact that, under the conditions of these experiments, the organic test compounds must serve as the sole sources of both the nitrogen and the carbon required for growth. When the glycine concentration is raised to 0.01 M, growth is somewhat inhibited, and complete inhibition is observed at a level of 0.03 M.

The availability of a bacterial strain which grows in a simple synthetic medium, and which shows a pronounced nutritional preference for a peptide rather than a mixture of the amino acids of which the peptide is composed, invited the examination of the influence of antibacterial agents on its growth. If antibiotics such as penicillin affected the nitrogen metabolism of this organism, this effect might be different depending on whether the organism was grown in the presence of a peptide or of the component amino acids. The data in Fig. 2 describe the result of a representative experiment in which strain SF was inoculated into media containing either L-leucylglycine or a mixture of L-leucine and glycine, together with increasing quantities of penicillin G sodium salt (Pfizer). In these tests, freshly prepared filter-sterilized solutions of penicillin were added, immediately before inoculation, to the autoclave-sterilized media.

It will be seen from Fig. 2 that while 500 Oxford units (O. u.) per ml inhibited bacterial growth only slightly in the presence of the peptide, smaller quantities of penicillin (1–10 O. u. per ml) exerted a striking inhibitory effect when the peptide was replaced by the mixture of the component amino acids.

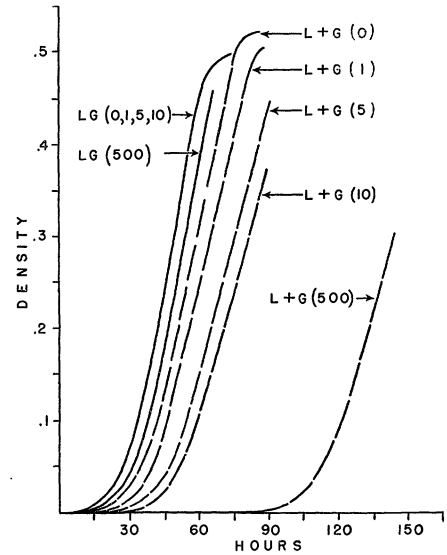

FIG. 2. Representative growth curves showing the effect of varying concentrations of penicillin on the growth of strain SF in the presence of 0.005 M L-leucylglycine (LG, solid lines) and of a mixture of 0.005 M L-leucine and 0.005 M glycine (L + G, dash lines). The concentration of penicillin, in Oxford units per ml, is given in parentheses for each curve. To facilitate inspection of the curves, the observed density readings (2-log galvanometer reading) have been omitted.

Information as to the mechanism of the inhibitory action of penicillin on the growth of strain SF in the presence of the amino acid mixture was provided by experiments in which each of the two amino acids served as the sole organic nutrient in the medium. When L-leucine was present, some inhibition was observed at 500 O. u. per ml, but not at the lower levels of penicillin employed. On the other hand, bacterial growth in the presence of glycine was markedly inhibited by 5 O. u. of penicillin per ml. This indicates that the inhibitory action of penicillin on the growth of strain SF in a medium containing L-leucine and glycine may be attributed largely to its interference with the utilization of glycine.

In view of the known bactericidal action of penicillin, it was necessary to determine the extent to which the number of viable cells originally present in the inoculum had been reduced during the early hours of the experiments in which marked inhibition of growth was observed. Plate counts were performed, therefore, on aliquots removed from the tubes at 5 hr and 10 hr after inoculation and, as will be seen from Table 1, a marked bactericidal effect in the presence of L-leucylglycine was evident with

500 O. u. of penicillin, but not at the lower penicillin levels. With the mixture of L-leucine and glycine, however, some killing was observed at 10 O. u. per ml (after 10 hr), but no appreciable effect was noted at 5 O. u. per ml. It may be concluded, therefore, that the inhibition of bacterial growth by 1–10 O. u. of penicillin per ml cannot be attributed to a bactericidal effect, but appears rather to be due to an inhibition of the utilization of glycine for growth.

In view of these results, the hypothesis may be offered that the assimilation of glycine by strain SF involves its prior incorporation into a peptide, and that the bacteriostatic action of penicillin is due to an inhibition of peptide synthesis. If this interpretation is correct, it may be hoped that further studies of the action of penicillin, and of other peptide antibiotics, on the utilization of amino acids and peptides by strain SF, as well as by other microorganisms, may serve to elucidate important aspects of the biosynthesis of peptide bonds.

TABLE 1

EFFECT OF PENICILLIN ON VIABLE CELL COUNT OF STRAIN SF

Penicillin (Oxford units per ml)	Total viable cells per tube × 10^{-8} *			
	L-Leucylglycine		L-Leucine + glycine	
	5 hr	10 hr	5 hr	10 hr
0	1.1	1.7	0.59	0.97
1	3.8	1.9	0.93	1.8
5	0.82	1.4	0.47	0.64
10	1.1	1.1	0.62	0.32
500	0.52	0.17	0.13	0.0075

* Immediately after inoculation, each tube contained approximately 1.1×10^8 cells.

Clearly, if incorporation into a peptide is an obligatory step in the assimilation of glycine by strain SF, it must, of necessity, be coupled to a process that will provide the requisite energy for peptide synthesis. Such an energy-yielding process may well be associated with the dissimilation of the carbon skeleton of the amino acids, and especially that of leucine. As has been shown by Gale and Taylor (*1*, *2*), the assimilation of some amino acids, notably glutamic acid, by *Staphylococcus aureus* requires the concomitant occurrence of energy-yielding reactions. These authors also made the significant discovery that penicillin impairs the ability of this organism to assimilate glutamic acid.

It is of interest to recall the recent observation (*4*) that the dipeptide glycyl-L-leucine exerts a bacteriostatic effect on the growth of a *leucineless* mutant of *Escherichia coli*, despite the fact that the peptide can be utilized by the mutant as a source of leucine. Since the same bacteriostatic effect of the peptide on the growth of the mutant was observed in the presence and in the absence of added L-leucine, it was concluded that glycyl-L-leucine interferes with the bacterial utilization of this amino acid. These results offer an example of a simple peptide that inhibits a key metabolic reaction of a microorganism in a manner that is strikingly similar to the effect of penicillin on the metabolism of strain SF.

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Effect of 2,4,5-Trichlorophenoxyacetic Acid on Ripening of Apples and Peaches

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Blondeau and Crane (*1*) have reported that the maturation of Lob Injir (Calimyrna) fig fruits in California has been advanced considerably as a result of spraying unpollinated but pollen-receptive synconia with solutions of 2,4,5-trichlorophenoxyacetic acid (2,4,5-T). In previously reported experiments on ripening of harvested banana, apple, and pear fruits, Marth and Mitchell (*2*, *3*) found 2,4-dichlorophenoxyacetic acid to be very active. Of the few compounds tested in the present experiments, 2,4,5-T has proved to be among the most active in bringing about the fruit-ripening response of apples and peaches on the tree.

On August 23, 1948, individual tagged fruits of Rome Beauty apples and adjacent spur leaves were sprayed at the Plant Industry Station, Beltsville, Maryland, with aqueous solutions of 2,4,5-T at 10-, 100-, and 200-ppm concentrations. Fruits that received either the 100- or the 200-ppm spray concentration developed red coloration and were maturing rapidly by September 13. This same stage of maturity on unsprayed fruits was not attained until one month later, October 12, the usual harvest date for this variety. At 10-ppm concentration, the spray had no observable effect. Measurements on fruit softening were made on September 27 with the aid of a fruit pressure tester. At this time the untreated fruits showed an average pressure reading of 25.9 lb, whereas the fruits sprayed with 10-, 100-, and 200-ppm concentrations of 2,4,5-T tested 24.8, 19.8, and 18.9 lb, respectively.

The spur leaves of Rome Beauty trees that received the 200-ppm concentration were burned by the spray. New shoot growth produced in the spring of 1949 was deformed and flowers were late in opening as a result of this treatment. No injury was found in the case of the 10- and 100-ppm sprays, either on foliage present at the time of spraying or on that produced the following year.

In 1949, three early summer varieties of apples also showed a ripening effect from spray applications of 2,4,5-T to foliage and fruit. The Close variety, which matures normally during the last week of June at Beltsville, Maryland, was induced to ripen 5–7 days early by spray applications of either 50- or 100-ppm concentration applied on June 10. The fruits on branches of Duchess apple that received a 50-ppm spray on June 10 colored and