

and then centrifuged. The supernatant liquid is added to a suspension of 10 g gliadin (prepared from wheat gluten by extracting it with alcohol of 70 per cent.<sup>4</sup>) in 200 cc of water. The mixture is kept for one hour under toluene at 37°, then adjusted to pH 7-8 by addition of 0.1 N ammonia, and left over night at 37°. The next day the pH of the mixture is again brought to 7-8 by addition of ammonia. After being kept at 37° for an additional 4-5 hours, the mixture is heated for 15 minutes in a boiling water bath and is then filtered over night in the ice box. The filtrate is concentrated *in vacuo* to a volume of 50-70 cc. It is turbid at first, presumably due to presence of emulsified toluene, and becomes clear during the concentration. In order to avoid foaming during the concentration step, alcohol is constantly added dropwise from a dropping funnel whose tap has been opened to a suitable degree. To the concentrated solution absolute alcohol is added until a strong turbidity is produced. The turbid solution is then poured with stirring into ten volumes of absolute alcohol. The white flocculent precipitate is allowed to settle and is then filtered by suction, washed with absolute alcohol and dried in a vacuum desiccator over sulphuric acid. Yield, 4-4.5. The peptone is free of ammonium salts. On acid hydrolysis it yields 4.61-4.74 per cent. ammonia, which corresponds to a total glutamine content of about 40 per cent. if the small asparagine content of gliadin is neglected).

Gliadin peptone in a concentration of 20-40 mg per cent. effectively replaced glutamine as a growth factor for hemolytic streptococci in the medium of McIlwain *et al.* It has also been found that the gliadin peptone effectively replaces Bacto-peptone "Difco" employed by McIlwain *et al.* The following medium, which is entirely suitable for the growth of the streptococcus, has been adopted by us: glucose 0.5 per cent.; gliadin peptone 1.0 per cent.; NaCl 0.03 per cent.; Na<sub>2</sub>HPO<sub>4</sub> 12 H<sub>2</sub>O 0.5 per cent.; KH<sub>2</sub>PO<sub>4</sub> 0.035 per cent.; MgSO<sub>4</sub> 7 H<sub>2</sub>O 0.03 per cent. The substances should be dissolved in the order given, and the solution heated to boiling for ten minutes, filtered and then autoclaved at 15 pounds for 30 minutes. pH = 7.6. After cooling, the following sterile ingredients are added to the basal mixture: riboflavine, calcium pantothenate and thiamine in amounts of 100 micrograms per cent. The results of a typical experiment are shown in Table 1.

It has also been found by us<sup>5</sup> that  $\alpha$ -methylamide and  $\alpha$ -ethylamide of glutamic acid<sup>6</sup> fail to act as growth factors for hemolytic streptococci in the

<sup>4</sup> Th. B. Osborne and E. Strauss, Abderhalden's "Handbuch der biologischen Arbeitsmethoden," I: 8, 437, 1922.

<sup>5</sup> With collaboration of Mrs. J. Storch-Levy.

<sup>6</sup> N. Lichtenstein, *Jour. Am. Chem. Soc.*, 64: 1021, 1942.

TABLE 1  
GROWTH OF STREPTOCOCCUS HAEMOLYTICUS "RICHARDS."  
GROWTH AFTER 24 HOURS AT 37°

Medium	Photometer reading
McIlwain's medium without glutamine	97*
" " + glutamine	0.03 mg† 64
" " + " "	0.1 " 58
" " + gliadin peptone‡	0.2 " 96.5
" " + " "	1.0 " 75
" " + " "	2.0 " 48
" " + " "	4.0 " 45
Gliadin peptone medium	54

\* Growth was measured photometrically. The photometer reading of tubes containing water was adjusted to 100. Sterile medium then gave a reading of 97. Increasing growth is reflected by a decreasing reading.

† Amounts per 10 cc medium.

‡ The pure gliadin peptone solution was autoclaved at 15 pounds for 30 minutes.

medium of McIlwain *et al.* McIlwain<sup>7</sup> has previously shown that N-acetylglutamine and a number of glutamine containing dipeptides (leucylglutamine, cystearylglutamine, glutaminyglycine, glutaminylcysteine and glutaminyglutamic acid) fail to replace glutamine as a growth factor for hemolytic streptococci. The results obtained with our peptone indicate therefore either that it contains free glutamine or that it contains peptides of glutamine which can be split hydrolytically by the proteases of the streptococci.

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#### ESTIMATION OF CATHEPSIN ACTIVITY

RECENTLY Plentl and Page,<sup>1</sup> in a study of renin preparations, have stated that the hydrolysis of benzoylargininamide was not followed quantitatively by an increase in titratable carboxyl groups since, with the concentrations of protein required, the large amount of precipitate formed during the titration masked the color change of the indicator. The procedure referred to<sup>2</sup> is that of Grassmann and Heyde.<sup>3</sup> Since this method has become so important in the kinetics of proteinase action,<sup>4</sup> and in order that others might not be discouraged, it seems worth while to point out that the above-mentioned difficulty was encountered in a similar case and circumvented not long ago.<sup>5</sup>

<sup>7</sup> H. McIlwain, *Biochem. Jour.*, 33: 1942, 1933.

<sup>1</sup> A. A. Plentl and I. H. Page, *Jour. Biol. Chem.*, 155: 368, 1944.

<sup>2</sup> K. Hofmann and N. Bergmann, *Jour. Biol. Chem.*, 130: 81, 1939.

<sup>3</sup> W. Grassmann and W. Heyde, *Zeits. Physiol. Chem.*, 183: 32, 1929.

<sup>4</sup> G. W. Irving, J. S. Fruton and Max Bergmann, *Jour. Biol. Chem.*, 138: 231, 1941.

<sup>5</sup> F. M. Uber and A. D. McLaren, *Jour. Biol. Chem.*, 141: 234, 1941.

The Grassmann and Heyde method calls for the titration of *alpha*-aminocarboxylic substances with alcoholic KOH to an endpoint with thymolphthalein comparable in depth of color to that of a dilute, ammoniacal  $\text{CuCl}_2$  solution. If to the color standard there is added a small amount of freshly precipitated  $\text{BaSO}_4$ , a turbidity is produced similar to that result-

ing from the precipitation of enzyme in the titration mixture. With this procedure, and a white box illuminated by a "daylight" bulb, data suitable for the calculation of reaction constants for trypsin were acquired.<sup>5</sup>

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## DISCUSSION

### BIOLOGY IN THE PREMEDICAL CURRICULUM

MANY colleges and universities are in the process of reevaluating their aims and teaching methods and of revising their curricula. The preprofessional curricula are necessarily affected by these changes. As far as the premedical curriculum is concerned, it is fortunate that medical schools and colleges seem to be in agreement on the basic principles on which this program should be based. Spokesmen of the American Association of Medical Colleges<sup>1,2</sup> as well as individual administrators and teachers of medical schools have repeatedly stressed the necessity of a broad cultural background rather than a narrow and specialized training in sciences, as the basis for the medical school and medical profession. For the medical student, an appreciation of arts and literature and an insight into social institutions and their history is deemed just as necessary as a thorough grounding in the natural sciences. The foundation for these qualifications must be laid in the college, which means that in a premedical curriculum the humanities, social sciences and natural sciences should be well balanced. Since the liberal colleges stand for the same general principle, there should be no difficulty in fitting the premedical program into the framework of an A.B. curriculum, by making allowance for the special needs of premedical students and by giving flexibility to the program of those who do not intend to take the A.B. degree. The difficulties become apparent when details are considered. Many difficulties arise from the fact that the entrance requirements of different medical schools differ widely. A more serious dilemma faces the sciences. If the principle outlined above is to be adopted sincerely, then premedical students should not be encouraged to specialize in natural sciences except in the case of those students who are especially gifted for them. On the other hand, those aspects of physics, chemistry and biology which are of importance for medicine grow steadily; so much so that, for instance, soon the need for a second year of physics may become urgent. The situation is aggravated by the prospect that in the future many,

if not the majority of all premedical students will not stay for more than three years in college. This means that the natural sciences are forced to accomplish more in a shorter time. It is urgently necessary that we scrutinize carefully and rigorously the contents of the courses which we offer to premedical students, and that we improve the efficiency of our teaching methods.

Last summer a small group of biologists and two members of medical schools met at the Marine Biological Laboratory in Woods Hole for an informal discussion of some of these problems as far as they concern the role of biology in the premedical curriculum. Those present were Ph. Armstrong, Syracuse University Medical School; E. Ball, Harvard University Medical School; L. V. Heilbrunn, Department of Zoology, University of Pennsylvania; R. Kempton, Department of Zoology, Vassar College; D. Marsland, Department of Biology, New York University; A. K. Parpart, Department of Biology, Princeton University; A. W. Pollister, Department of Zoology, Columbia University; W. R. Taylor, Department of Botany, University of Michigan, and the writer. It was felt by the group that some of the conclusions reached in this conference might be of a more general interest and might serve as a basis for further discussion. It should be stated that the participants expressed their personal opinions and not those of any organizations or institutions. Furthermore, this report is based on the spontaneous discussion which developed during the conference and makes no claim to cover the ground adequately. The following ten points present the edited and somewhat enlarged protocol of the meeting.

(1) The entrance requirements of the different medical schools differ widely from each other (see the tabulation of Swett<sup>3</sup>). The premedical curricula of colleges are even more diversified, due to local conditions and traditions, and because the colleges have the responsibility of preparing premedical students for more than one medical school. We recognized fully that a certain degree of diversity is desirable and also inevitable, but it was felt that at present there exists too much variation. As a result the stu-

<sup>1</sup> W. C. Rappleye, *Jour. Assoc. Am. Med. Coll.*, 15: 221-227, 1940.

<sup>2</sup> F. C. Zapffe, *ibid.*, 15: 228-234, 1940.

<sup>3</sup> F. H. Swett, *ibid.*, 15: 385-386, 1940.