

became desert, the wind-blown sand from the region south of the Mediterranean should begin to make its appearance. This wind-blown sand is abundant in the sediments forming to-day in the deep basins of the Mediterranean.²²

Less vague, however, would be the record left by the activity of explosive volcanoes. A considerable number of these ash falls in the upper part of the post-Pleistocene column of sediments should be correlatable with human history. Earthquakes, too, should have caused submarine mud slumps that threw into suspension much sediment which settled as widespread blankets²³ of distinctive sediment. Such a blanket of sediment should show a gradation in grain size due to the differential settling rates of the constituent particles thus thrown temporarily into suspension.

It seems to me, therefore, that long cores of the sediments in the deep basins of the Mediterranean would probably reveal an extraordinarily rich and varied record in a locality critical not only by reason of the unusual configuration of the basin but also by reason of the wealth of information that is already available from the long historic records, the archeology and the Pleistocene and post-Pleistocene geology.

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TYROSINE DETERMINATIONS¹

THE author was recently engaged in a study of egg albumin in which it was necessary to run tyrosine determinations on small quantities of the protein. Since the method of Folin and Marenzi² requires 100 milligrams for each determination, and that of Lugg,³ approximately 50 milligrams of protein, it was decided to use Lugg's method. It is for the purpose of discussing modifications of this method that this paper is presented.

In the method under discussion, that of Lugg, the 50 milligram sample of ovalbumin is hydrolyzed in alkali as recommended by Folin and Marenzi, acidified, centrifuged and a 3 ml aliquot diluted to 5 ml with 5 and 1 normal H_2SO_4 in such proportions that the 5 ml solution will be at a pH of 0.3 (from titration of a separate aliquot using brilliant cresyl blue as an indicator). It is common knowledge that no efficient indicator is available at this pH range and that the glass and quinhydrone electrodes are extremely inaccurate for this determination. In search of a simpler

method of bringing the pH of the standard and unknown solutions to the same and required value it was discovered that one could very well utilize the normality of the test solutions with respect to H_2SO_4 . When the 5 ml test solution is made 1 normal H_2SO_4 the buffer action of the amino acids in the solution and the repression of the ionization of the strong electrolyte (H_2SO_4), results in a pH of approximately 1. Block⁴ states that a pH of 2.5 is also satisfactory. A series of analyses using the modification outlined here resulted in the following values for the per cent. tyrosine in egg albumin:

3.77; 3.83; 3.85; 3.81; 3.77; 3.83; 3.84
Mean 3.81

These values are close to the figures reported in the recent literature. Bernhart⁵ reports 3.85 per cent. tyrosine in ovalbumin as determined by the method of Folin and Marenzi.

Further study of Lugg's method disclosed that another step could be modified for convenience. After the 5 ml test solution has been mercurated with a mixture of mercuric sulfate and chloride and made up to 25 ml, Lugg recommends that the sodium nitrite be added within an hour and compared colorimetrically with the standard (Millon reaction), since cloudiness may develop on longer standing and hinder the comparison. The following experiment was performed:

Six samples of egg albumin were treated simultaneously and in an entirely analogous manner according to the above discussed modification. After the solutions were made up to 25 ml, four samples were compared immediately, and the remaining two, twenty-four hours later. The results obtained from the two sets, in per cent. tyrosine present, are outlined below (the samples were previously treated in an oven for 24 hours at 110 C.).

Analyzed immediately	Analyzed 24 hours later
3.61	3.75
3.67	3.68
3.75	
3.83	
Mean 3.72 per cent.	3.72 per cent.

These data can be interpreted to indicate that the test solution, diluted to 25 ml, can remain at least 24 hours before it is compared colorimetrically with the standard without any appreciable decrease in value of the tyrosine present.

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²³ Fr. Nipkow, *Rev. d'Hydrologie*, 4 Année, No. 1/2: pp. 70-120, 1927.

¹ From the laboratory of Physiological Chemistry, University of Minnesota, Minneapolis.

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³ J. W. H. Lugg, *Biochem. Jour.*, 31: 1422, 1937.

⁴ R. J. Block, "The Determination of the Amino Acids," Burgess, Minneapolis, Minnesota, page 21, 1938.

⁵ F. Bernhart, Unpublished Ph.D. thesis, University of Minnesota, 1938.