of CO_2 and O_2 in the original sample are calculated from the changes in length of the gas column, and the volume of the bubble is calculated from the length of the gas column (40–100 mm.) and the cross-section area of the capillary (.0095 mm.^{2}). A complete analysis can be performed in 10–15 minutes.

After each analysis the apparatus is cleaned by drawing tap water and then cleaning solution through the capillary, rinsing with tap and distilled water.

Accuracy

The accuracy of analysis depends primarily on the amounts and kinds of gases in the sample. Other important factors, subject to a considerable degree of control, are (1) alteration of the sample through gaseous exchange with the analyzer fluid by diffusion; (2) changes in the water vapor tension in the bubble when in contact with solution of different osmotic pressure; (3) rate at which the bubble is drawn into the capillary; and (4) reading error. In actual practice, diffusion of gases is found to be the greatest source of error. If the composition of the bubble diverges only slightly from the gas tensions in the analyzer fluid, which is equilibrated with air, the error due to diffusion is negligible, but it increases as the composition of the sample diverges from that of air. This error was found to be considerably reduced by (1) using saturated aqueous LiCl for the analyzer and transference fluids; (2) using alkaline LiCl solution as the CO_2 absorbent; and (3) introducing the gas sample into the analyzer as rapidly as possible.

TABLE 1

Sample No.	Vol. of bubble	Known composition of gas mixture*			Composition as measured in analyzer		
		CO_2	O_2	N_2	CO_2	O_2	N_2 (by difference)
	(mm.^{3})	(%)	(%)	(%)	(%)	(%)	(%)
1	1.43				...	20.7	79.3
2	0.42	0.03	20.95	79.02	...	21.0	79.0
3	1.41		(air)		...	20.9	79.1
4	0.78				...	21.1	78.9
1	0.55				5.0	91.1	3.9
2	0.45	5.29	93.63	1.08	4.9	89.6	5.5
3	1.03				5.4	92.6	2.0
4	0.40				5.4	92.1	2.5
1	0.50				14.5	8.1	77.4
2	0.57	14.90	7.65	77.45	14.7	8.0	77.3
3	0.45				14.4	7.9	77.7

* Determined by means of the Haldane gas analysis apparatus.

Greater accuracy could be obtained by equilibrating the solutions with a gas mixture of composition approximating that of the sample to be analyzed. Under most favorable conditions the error of analysis is less than 0.3 per cent. Under less favorable conditions it increases but is not unreasonable considering the extremely small volumes of gases analyzed.

Typical analyses of air and known gas mixtures by this method are given in Table 1.

References

1. Harris, M., Berg, W. E., Whitaker, D. M., Twitty, V. C., and Blinks, L. R. *J. gen. Physiol.*, 1945, **28**, 225.
2. Hartridge, H. *J. sci. Instr.*, 1939, **16**, 317.
3. Krogh, A. *Skand. Arch. Physiol.*, 1908, **20**, 279.
4. Scholander, P. F. *Rev. sci. Instr.*, 1942, **13**, 264.

A New Method for the Purification of Arginase

C. Bertrand Thompson[1]

Division of Biochemistry, University of California Medical School, Berkeley

Many and varied studies on arginase can be and have been made on very crude solutions. With two exceptions, most preparations reported from 1924 (*3*) to 1944 (*2*) were essentially extractions in glycerol, with reduction in large volume of acetone. The exceptions were Edlbacher and Simons (*1*), who tried adsorption on alumina C and Willstätter kaolin, and Richards and Hellerman (*5*), who made repeated fractionations in acetone, ammonium sulfate, sodium salicylate, and sodium alizarin sulfonate, with frequent dialyses. These methods gave products which, although much purer than the usual solutions, were neither comparatively very active nor very pure.

The first reported systematic study of preparative methods is that of Mohamed and Greenberg (*4*). The procedure finally adopted by them consisted in extractions and fractional precipitations in sodium acetate, lead acetate, ammonium sulfate, adjustment to pH 8,

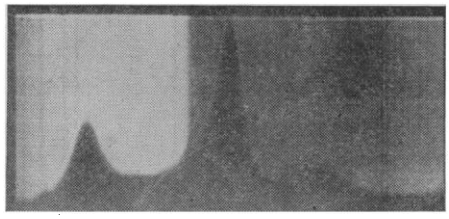

Fig. 1. Tiselius curve of purified arginase in pH 6.2 phosphate buffer μ0.10; 205 minutes at 5.8 volts/cm.; $U_P = 6.3 \times 10^{-5}$; $U_S = 2.3 \times 10^{-5}$. (Courtesy of Dr. C. H. Li, Institute of Biological Research, University of California.)

reduction in acetone, and solution in pH 7 phosphate buffer. The final product, a green-brown solution, showed by electrophoresis in the Tiselius apparatus the presence of three or four constituents. Catalase was a definite contaminant of the mixture. The problem remaining, therefore, was that of freeing the preparation of catalase and other proteins. It was

[1] The author wishes to thank the Division of Biochemistry, University of California Medical School, for the use of its laboratories and facilities, and Prof. D. M. Greenberg, Division of Biochemistry, for helpful suggestions and advice.

at this point that the present investigation was undertaken by the author.

The addition of manganese or cobalt salts, brought to pH 4, with the immediate addition of an excess of phosphate buffer of pH 9 cleared the solution, apparently by coprecipitation, of all colored substances, with loss of only 35–45 per cent of activity. Details of the method will be given elsewhere.

The Tiselius curve (Fig. 1) shows the presence of only two constituents, presumably only one besides the arginase. The arginase is found in the slow fraction.

The spectrophotometric curve (Fig. 2) of the cobalt-purified solution (A) and a solution before purification (B) shows a considerable drop in the peak at 412 mμ.

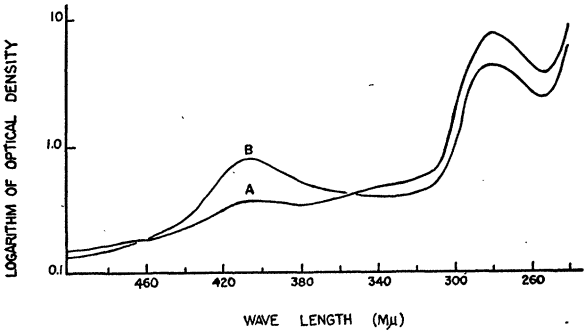

FIG. 2. Spectrophotometric curve of purified (A) and unpurified (B) arginase. (Courtesy of L. A. Strait, University of California Hospital.)

It may be considered established that arginase is a colorless enzyme, with the properties of an albumin. Further studies are under way to complete the purification and to increase the yield.

TABLE 1
EFFECT OF DIVALENT CATIONS

Salt	Clarification	Yield
Co^{++}	Excellent	Good
Mn^{++}	Good	"
Ni^{++}	"	"
Cd^{++}	"	Poor
Zn^{++}	"	"
Sr^{++}	Slight	Good
Ba^{++}	"	Fair
Ca^{++}	"	"
Mg^{++}	"	"
Pb^{++}	"	Poor

Another series of tests shows similar effects in varying degrees from the following divalent cations: barium, cadmium, calcium, lead, magnesium, nickel, strontium, and zinc. Cobalt, manganese, and nickel give the best combination of clarification and yield; cadmium and zinc give excellent clarification but less than half the yield; while barium, lead, magnesium, and strontium give comparatively slight clarification and widely varying yields (Table 1).

References

1. EDLBACHER, S., and SIMONS, S. *Z. physiol. Chem.*, 1927, **167**, 76.
2. HUNTER, A., and DOWNS, C. E. *J. biol. Chem.*, 1944, **155**, 173.
3. LEUTHARDT, F., and KOLLER, F. *Helv. Chim. Acta*, 1924, **17**, 1030.
4. MOHAMED, M., and GREENBERG, D. M. *Arch. Biochem.*, 1945, **8**, 349.
5. RICHARDS, M. M., and HELLERMAN, L. *J. biol. Chem.*, 1940, **134**, 237.

Letters to the Editor

On Filing Reprints

L. R. Richardson (*Science*, 1946, **104**, 181–182) explained a method of filing which he has found to be very satisfactory for a reprint collection. He recommended filing by author in large, 10- by 13-inch, open-ended manila envelopes, one author per envelope. There seem to be some objections to this system. These will be mentioned and another system proposed.

In the first place, many bookcases are not provided with sufficient clearance between shelves so that the 13-inch envelopes can be placed on end as suggested, and if they are, a lot of potential shelf space is lost. Secondly, a great percentage of reprints do not require envelopes of such size, but because of the occasional one of large format or the possible future one, extra room must be provided in all envelopes. Thirdly, many reprint collections contain only one or two papers by some authors and dozens by others. A manila envelope for a single paper might be considered extravagant both in money and space, while a prolific writer would require many envelopes. The last objection is that the reprints stand in a vertical position, which is not the best from the point of view of preservation, particularly for old papers.

After having tried several systems, the present writer has found one that is workable and conserving of space. The fundamental assumption is that the place of original publication rather than subject or author is the best basis for filing. This method does away with the difficulty of subject classification and with the problem of joint authorship—the best-known author's name does not always come first. Under this system papers are filed with others of the same size, since journals do not constantly change their format, and it is not necessary to provide unnecessary space for the occasional large paper as under the system proposed by Richardson.

Cardboard boxes with deep covers are used, to which are pasted labels listing the periodicals contained. Within the box, papers are filed by date of publication. Reprints