

Since there is no absolute measure of blood volume the important requirements in any clinical method are reproducibility and constancy of the normals. These requirements are fulfilled by the present method. In 42 determinations the normal range obtained corresponded closely to those reported by Storaasli and co-workers (2): total blood volume, 60–85 cc/kg; plasma volume, 34–60 cc/kg; and red cell volume, 26–40 cc/kg.

The method herein reported permits the rapid determination of blood volumes repeated at short intervals.

References

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The Oxidative Deamination of Serotonin and Other 3-(beta-Aminoethyl)-indoles by Monamine Oxidase and the Effect of These Compounds on the Deamination of Tyramine

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Recent interest in the vasoconstrictor element of blood serum which had led to the elucidation of the structure of Serotonin (1–3) has resulted in the synthesis of this compound (4) and other derivatives of 3-(beta-aminoethyl)-indole (5) in these laboratories. Blaschko has reported that Serotonin itself is deaminated oxidatively by guinea pig tissue (6).

In an effort to assess the possible oral activity of the Serotonin derivatives reported herein, it was thought worth while to investigate their deamination by monamine oxidase.

The method was essentially that employed by Beyer (7) utilizing manometric determination of the increase in oxygen uptake by guinea pig liver homogenate in the presence of the amines above the blank oxygen utilization. Cyanide was used as aldehyde fixative. Tyramine was run as a standard in each manometric determination. All compounds were subjected to deamination in the amount of 1.25×10^{-5} mole. Thus the theoretical oxygen uptake/atom of oxygen used was 140 mm³.

It will be seen in Table 1 that similarly to the findings in the deamination of substituted phenyl ethylamines (8) substitution of a single methyl group in the side chain of these compounds does not impair deamination (see compounds II, IX). Dimethyl, isopropyl, benzyl, or diethyl substitution of the aminoethyl nitrogen, precludes deamination by this system as does ethyl substitution of the beta carbon of this

TABLE 1

Compound	Name	O ₂ Uptake, mm ³
I	3-(beta-Aminoethyl)-indole (acetate)	165
II	3-(beta-N-Methylaminoethyl)-indole (hydrochloride)	160
III	3-(beta-N-Benzyl-N-methylaminoethyl)-indole (hydrochloride)	-10
IV	3-(beta-N-Isopropylaminoethyl)-indole (hydrochloride)	0
V	3-(beta-N-Benzylaminoethyl)-indole (hydrochloride)	-10
VI	3-(beta-Aminobutyl)-indole (hydrochloride)	0
VII	3-(beta-N,N-Diethylamino)ethyl indole (hydrochloride)	0
VIII	5-Hydroxy-3-(beta aminoethyl)-indole (Serotonin) (creatinine sulfate monohydrate)	152
IX	5-Hydroxy-3-(beta-N-methylaminoethyl)-indole (creatinine sulfate monohydrate)	158
X	5-Hydroxy-3-(beta-N,N-dimethylaminoethyl)-indole (creatinine sulfate monohydrate)	28
	Tyramine	140
	Creatinine sulfate	0

chain (see compounds X, IV, V, VII, and VI, respectively). The small oxygen uptake seen with compound X (5-hydroxy ring-substituted) when compared with other compounds not ring-substituted and in which the aminoethyl nitrogen atom is blocked (III, IV, V, VI, VII) would suggest possible oxidation of the ring hydroxyl group.

This cannot be confirmed in the comparison of compounds VIII and IX with I and II, respectively, since all these compounds show a greater oxygen uptake than does tyramine. It is possible that this excess oxygen consumption could signify some ring oxidation other than that of the 5-hydroxy group.

Creatinine sulfate (complexed with compounds VIII, IX, and X) was not oxidized when examined separately. The rate of deamination of the compounds that were oxidized, although not illustrated, is uniformly more rapid than that of tyramine.

Since certain of these compounds showed slight depression of the basal oxygen uptake of the homogenate, it was thought of interest to investigate inhibition of tyramine oxidation by some of those compounds not deaminated. This was done by adding the compound to be tested for inhibition to both control and tyramine containing vessels. The percentage of in-

TABLE 2
INHIBITION OF TYRAMINE OXIDATION
(TYRAMINE 0.0066 M FINAL)

Compound	Final concentration	Inhibition, %
III	0.0066 M	37.2
IV	0.0066 M	83.8
V	0.0066 M	35.2
VI	0.0066 M	65.0
VII	0.0066 M	89.3

hibition was calculated on the basis of the maximum rates during the first 30 min.

It may be seen in Table 2 that these compounds do in fact inhibit tyramine oxidation. Since the amounts required are in the same range as the concentration of tyramine, it is presumed that the inhibition is of a competitive nature.

On the basis of this study, it would appear likely that compounds III, IV, V, VII, and X would be more active orally than the remaining compounds, since they are not deaminated by liver tissue. It should be borne in mind, however, that Beyer and Stutzman (8) have shown that tyramine can be recovered apparently intact from urine after oral administration, showing that hepatic deamination may not always be operative *in vivo*. Their finding, however, does not preclude reamination of *p*-hydroxyphenyl acetic acid in the kidney.

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The Appearance of Amoebae Tracks in Cultures of *Dictyostelium discoideum*

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A recently published paper of Bonner (1) indicates his continuing interest in the formation of the slime trail of the pseudoplasmodium of *Dictyostelium discoideum*, a slime mold of the order Acrasiales in the Myxomycophyta. A footnote appearing in the paper speculates on the possibility of the pseudoplasmodium trail being formed by slime given off by the individual myxamoebae which make up the migrating pseudoplasmodium.

Prior to the appearance of Bonner's paper (1), in January 1953, it was found that the individual myxamoebae of a culture of *D. discoideum* were leaving tracks or trails on the substrate of a plate of glucose medium. The observation was made under illumination of low intensity. The trails were best seen when the microscope was focused down, giving the myxamoebae the appearance of black spots with a bright halo. The trails appeared grayish. The culture on which this phenomenon was observed had been subcultured from an original collection of the organism made by D. D. Perkins of Stanford University. The pseudoplasmodia of this strain had completely lost their ability to migrate. Pseudoplasmodia were formed at centers of aggregation and sorocarps developed directly from the sedentary pseudoplasmodia. The slime mold in this

original observation was grown in a two-membered culture with *Bacillus subtilis*.

Recently the work of January 1953 was repeated.¹ A culture of a motile strain of *D. discoideum* was secured from Ralph Emerson of the University of California. Both the motile and the previously used non-motile strains of the slime mold were cultured on four different media: (a) Emerson's yeast extract (2), one-half strength; (b) a modification of the Emerson medium in which glucose was substituted for starch; (c) cow dung agar; and (d) tap water agar.

It was found that the myxamoebae of both strains left trails on the various media used. The tracks were found about 5 days following the inoculation of the culture. In repeating the experiment *Escherichia coli* was used as the second member of the culture. It was observed that the frequency of the appearance of the tracks was positively correlated with the richness of the medium used. Glucose and yeast extract media showed many trails were formed which were well-defined when examined microscopically in reduced light at 100×. The tracks were only occasionally found on cow dung and tap water media.

Shown below are two photomicrographs (Figs. 1A

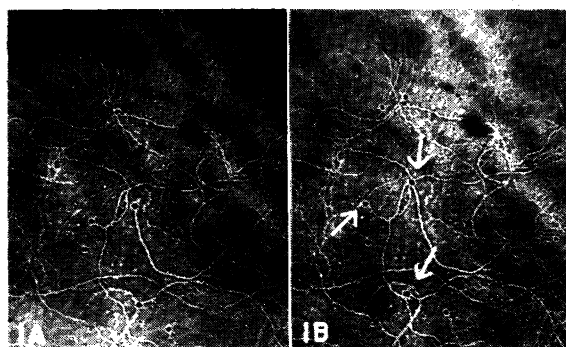


FIG. 1. A, *Dictyostelium discoideum* vegetative myxamoebae on yeast extract medium showing tracks. 100×; B, The same culture as 1A photographed 20 min later. Note the extensions of the tracks produced during the interval (see arrows). 100×.

and 1B) which were taken of a culture of *D. discoideum* grown on yeast extract medium. Figure 1B was photographed 20 min after Fig. 1A. The tracks or trails are clearly shown in the photographs. Close examination of the figures will show that the myxamoebae, which appear as black spots on the plates, have moved to new locations during the time interval and the extension of the trails is clearly seen.

Attempts to show that the slime trail of the pseudoplasmodium and the tracks of the myxamoebae are the same or similar substances have been unsuccessful to date. A 0.01% aqueous solution of Congo Red deposits some stain on the slime trail, but does not stain the myxamoebae tracks. The myxamoebae tracks disappear in the aqueous solution, but reappear in 10 or 15 min, presumably when the water of the staining

¹ The original observations and the repetition of this work were conducted in the laboratories of R. M. Page of the Department of Biological Sciences, Stanford University, California.