

However, it would seem that structural considerations other than -SH binding capacity are involved in alloxan action since the closely related, but non-diabetogenic compound, alloxanic acid, is the more potent -SH inhibitor. If one accepts the figures given for the rapid conversion of alloxan to alloxanic acid in blood plasma⁷ as representing the rate of alloxan breakdown after intravenous injection, then the animal contains primarily alloxanic acid in a matter of minutes. It is logical to suppose that a certain portion of the alloxan exerts its action before this conversion is complete. The same possibility exists with regard to the action of alloxan on isolated enzyme systems. It is, however, difficult to interpret many of the early *in vitro* studies because they were carried out at neutral pH's where alloxanic acid could have been responsible for the inhibition observed. While the introduction of controls in the form of aged neutral solutions of alloxan has given validity to certain recent studies, there has been no attempt to assign any background inhibition to a specific degradation compound such as alloxanic acid. The possibility that alloxanic acid may have a synergistic action by creating an -SH deficiency exists.

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CLARKE T. GRAY
MARCUS S. BROOKE
JOHN C. GERHART

Department of Bacteriology and Immunology,
and the Leonard Wood Memorial Laboratory,
Harvard Medical School, Boston.

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Paper Chromatography of Corticosteroids at Room Temperature

THE numerous paper chromatographic methods¹⁻³ so far presented for the separation of corticosteroids all have limitations; our results may therefore be of interest. We have found that by impregnating the papers with a 90 per cent methanol solution we could separate the corticosteroids rapidly at room temperature (16-20°C.). The method also works for prednisone and prednisolone (Table 1). Furthermore, it is suitable for quantitative and semi-quantitative determinations.

1-200 µgm of the steroids, dissolved in methanol, was spotted on to strips 2 cm. wide prepared from 45 cm. × 4 or 15 cm. Schleicher-Schüll 2043/b papers. The strips were then impregnated by drawing them through a 90 per cent methanol solution, leaving a dry space, 0.5-1 cm. wide, at the starting line. The excess liquid was pressed out between two layers of filter paper and the strips were immediately placed in the apparatus.

Two sizes of glass chamber were employed, depending on the size of the paper. Ten minutes before use, porcelain cups containing wads of paper were placed in each chamber. One cup was filled with the organic

Table 1. R_F OF CERTAIN STEROIDS AFTER IMPREGNATION IN 90 PER CENT METHANOL Solvent System*

Steroid	1		2		3	4	5	6
	R_F	R_{FC}^\dagger	R_F	R_{FC}^\dagger	R_F	R_F		
Hydrocortisone	0.24	0.55	0.09	0.46	0.12	0.39	0.16	0.49
Aldosterone	0.34	0.80	0.13	0.71				
Cortisone	0.40	1.0	0.17	1.0	0.20	0.56	0.37	0.64
Corticosterone	0.60		0.48		0.51	0.74	0.71	0.80
Reichstein's S compound	0.63		0.49		0.54	0.76	0.75	0.84
11-dehydrocorticosterone	0.68		0.66		0.67	0.85	0.84	0.90
11-deoxycorticosterone	0.81		0.90		0.85	0.96	0.98	0.98
Prednisolone	0.20	0.50	0.05	0.25				
Prednisone	0.34	0.82	0.13	0.72				

* Solvent mixture: 1, toluene-ethanol-petroleum ether-water (2:6:1:3); 2, Bush system⁴ toluene-methanol-water (10:7:3); 3, Bush² B 2 system; 4, Pechet⁵ No. 1; 5 Pechet No. 2; 6 Pechet No. 6.

[†] R_F values related to cortisone after 16 hours of development.

phase, the other with 90 per cent methanol solution. This served to saturate the atmosphere in the chambers. The small chambers were also lined with a piece of filter paper 20 cm. × 20 cm. saturated with the aqueous phase.

The organic and aqueous phases were prepared from a mixture of toluene-ethanol-petroleum ether-water (2:6:1:3). The mixture was allowed to stand for 18 hours and after separation the organic phase was filtered.

Development was carried out for 2-2.5 hr., during which time the front sank 30 cm. The cylinder was covered with a glass plate and the top and bottom were sealed with a glycerine-starch paste.

Under similar experimental conditions good separations were obtained with Bush's⁴ solvent mixture (toluene-methanol-water, 10:7:3), Bush's B2 system, and Pechet's⁵ solvents, Nos. 1, 3 and 6.

An alkaline solution of tetrazolium blue and 15 per cent phosphoric acid were most frequently used for detection.

Further experiments are in progress.

J. TUZSON

State Institute of Public Health
and First Medical Clinic,
University Medical School,
Budapest.

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Labiality of Fibrinolysin

FEARNLEY'S method for estimating fibrinolytic activity of normal blood¹ is based on the fact that the clot formed by adding thrombin to blood diluted 1 : 10 with phosphate buffer will lyse on incubation. This lytic activity may be lost if the specimen is permitted to stand for hours or even minutes at room temperature. Thus Fearnley and others caution that specimens must be kept in an ice bath and preferably collected in pre-chilled vessels. In contrast, the techniques used in our studies of fibrinolysis induced by nicotinic acid² employing essentially undiluted plasma and the 'thrombelastograph'³ fail to show spontaneous lysis of normal plasma. The lytic activity induced by parenterally administered nicotinic acid is clearly demonstrable by our methods, and is not destroyed by many hours storage at room temperature or even two weeks at 5° C.

When a 10 : 1 phosphate-diluted normal blood specimen is observed in the thrombelastograph, the