are, however, no strong grounds for expecting the same proportion in the rarer groups; and until we know more, perhaps as many as one in five of the rare chromosomes *CdE* should be regarded as $C^{w}dE$.

Mrs. "O's" blood groups are: *O, H+ve, $C^{w}dE$/cde, MsNS, P, Le(a−b+), Lu(a−), kk, Jk(a+)*.

We should like to express our thanks to Lieut.-Col. R. H. Malone, director of these laboratories, for permission to publish this communication, to Dr. R. R. Race for his criticism, to Prof. R. A. Fisher for his advice on frequencies, and to Miss E. W. Ikin, Blood Group Reference Laboratory, Lister Institute, London, for confirmation of our findings.

Note added in proof. Since submitting this communication for publication, the *Rh* chromosome *CdE* (R_y) has been proved to exist in two other families; see Grove-Rasmussen, M., *et al.*, *Ann. Eug.*, **16**, Pt. 2, 131 (1951); and Sussman, L. N., and Wald, N., *Amer. J. Hum. Genet.*, **2**, 85 (1950).

I. Dunsford
P. Aspinall

National Blood Transfusion Service,
Sheffield.

[1] Fisher, R. A., and Race, R. R., *Nature*, **157**, 48 (1946).
[2] van der Bosch, C., *Nature*, **162**, 781 (1948).
[3] Wiener, A. S., *Nature*, **162**, 735 (1948).
[4] Johnson, A., *Ann. Eug.*, **15**, 159 (1949).
[5] Race, R. R., Mourant, A. E., and Callender, S., *Nature*, **157**, 410 (1946).
[6] Stratton, F., *Nature*, **158**, 25 (1946).

Relative Importance of Free α- and ε-Amino Groups for the Biological Activity of the Growth Hormone

The biological activity of growth hormone[1] is found to be greatly reduced following treatment with acetic anhydride, which is known to acetylate protein amino groups specifically[2]. The relative importance of free α- and ε-amino groups has not hitherto been investigated. Purified growth hormone has now been acetylated according to the conditions of Li and Evans[1], but with smaller amounts of acetic anhydride so as to avoid complete inactivation.

The fluorodinitrobenzene technique of Sanger[3] was applied to untreated growth hormone in the first instance. The α-dinitrophenyl amino-acids liberated by acid hydrolysis were extracted with ether, and, after passage through a silica gel column, were identified by paper chromatography as dinitrophenyl-alanine and phenylalanine, in accordance with the results obtained by Li and Porter[4] in a preliminary study of the terminal amino groups of growth hormone. Spectrophotometric determination of the α-dinitrophenyl amino-acids was carried out on the eluate from the column. The ε-dinitrophenyl lysine hydrochloride remaining in the ether-extracted hydrolysate was estimated spectrophotometrically after passage through an acidic silica gel column.

Data for Acetylated Growth-Hormone Preparations

Acetic anhydride: moles per mole of growth hormone	Residual free amino groups per mole of growth hormone*		Growth-promoting potency, % of original potency
	Alanine + phenylalanine	Lysine (ε)	
—	1·4	19	(100)
120	<0·1	15·5	53
300	<0·1	13·5	26
2,700	<0·1	9·5	11

* Not corrected for losses in hydrolysis, etc.

When acetylation was performed with a relatively small amount of acetic anhydride, no α-dinitrophenyl amino-acids could be detected following fluorodinitrobenzene treatment of the acetylated product (see table). There was, however, only a partial reduction in the ε-dinitrophenyl lysine content of the hydrolysate and only partial loss of growth-promoting activity (assayed in intact female rats[5]). With larger quantities of acetic anhydride, there was a marked lowering of growth-promoting activity concomitant with increased acetylation of ε-amino groups, as shown by the ε-dinitrophenyl lysine determinations (see table). It thus appears that the growth-promoting activity of the growth hormone depends on the integrity of the ε-amino groups, but not on that of the terminal α-amino groups. The acetylated products were assayed for diabetogenic activity in the intact cat, as will be reported elsewhere. The results indicated that the diabetogenic activity of growth hormone also depends on the presence of ε-amino groups, but not on that of the α-amino groups.

The finding that acetic anhydride can be employed to acetylate selectively the free α-amino groups of growth hormone may be applicable to other proteins possessing both free α-amino groups and free ε-amino groups.

I am indebted to the Beit Memorial Trust for the award of a fellowship, to Dr. F. Sanger for advice on the use of his technique, and to Prof. F. G. Young for helpful advice and criticism.

E. Reid

School of Biochemistry,
University of Cambridge.
July 19.

[1] Li, C. H., and Evans, H. M., "Recent Progress in Hormone Research" (edit. G. Pincus), chap. 1, Vol. 3 (New York: Academic Press, Inc., 1948).
[2] Fraenkel-Conrat, J., and Fraenkel-Conrat, H., *Biochim. et Biophys. Acta*, **5**, 89 (1950).
[3] Sanger, F., *Biochem. J.*, **39**, 507 (1945).
[4] Li, C. H., and Porter, R. R. (unpublished experiment).
[5] Cotes, P. M., Reid, E., and Young, F. G., *J. Endocrinol.*, **6**, xiv (1949).

Bacterial Enzyme Preparations oxidizing Inositol and their Inhibition by Colchicine

Previous studies in this laboratory have brought to light a number of regularities governing the oxidation of isomers of the inositol group by resting *Acetobacter suboxydans*[1,2]. If the hexahydroxy *cyclo*hexanes are formulated as existing in the chair configuration, the minimum steric requirements for the oxidation of the various isomers by the resting cells are described in the statement that only those hydroxyl groups are oxidized that are situated in a polar plane.

The study of many aspects of these oxidative reactions required enzyme preparations in the cell-free state[3]. A thick suspension in 1/15 *M* phosphate buffer (*p*H 6·4) of the organisms, grown and harvested as described previously[2], was passed through a bacterial mill (Unicam, Cambridge). About one-half of the cells was broken. After centrifugation of the mixture (30 min., 2,000 *g*) and re-extraction of the sediment with buffer on a vibrator (60 vibrations per sec.) for 12 hr., followed by an intermediate centrifugation cycle at 11,000 *g* (1 hr.), the combined supernatants were finally centrifuged at 20,000 *g* for 2 hr. The reddish-brown pellet was suspended in phosphate buffer of *p*H 6·4. All operations were carried out in the cold.