by 70 per cent; DL-N-ethylleucine that of DL-leucine by 80 per cent; DL-N-hydroxyethylvaline that of DL-valine by 80 per cent; and DL-N-cyclohexylvaline that of DL-valine by 65 per cent. It was found in some cases that derivatives of one amino-acid inhibited the oxidation of other amino-acids more than that of the parent compound. For example, DL-N-ethylvaline inhibited the oxidation of DL-leucine by 45 per cent, whereas the oxidation of DL-valine was inhibited by 15 per cent only.

The oxidative deaminations of DL-valine, DL-leucine and DL-phenylalanine were also 23–30 per cent inhibited by DL-N-ethylleucylglycine.

The experiments were carried out in a Warburg apparatus at 38° by the usual technique. The *p*H varied between 8·1 and 8·5 (pyrophosphate buffer, 0·06 *M*). Aqueous extracts (usually 1 ml. per reaction mixture) of acetone-dried pig kidney preparations were used[1]. The final concentration of the DL-amino-acids was 0·008 *M* and that of the derivatives 0·04 *M*. The total volume of the reaction mixture was 2·5 ml. Oxygen uptake was measured after 60 min.

None of the amino-acid derivatives mentioned above was attacked by the enzyme to any significant extent.

It is known that the effects of crude enzyme preparations as used by us are the results of the combined activities of D-amino-acid oxidase proper and catalase[2]. The apparent inhibitions described above might therefore represent an increase in the rate of catalase action rather than an inhibition of the oxidase. We therefore tested the action of our enzyme extracts on hydrogen peroxide in separate experiments. The addition of either DL-N-ethylleucine, DL-leucinamide, DL-leucylglycylglycine or glycyl-DL-leucine had no influence on the catalase activity measured. Hence it may be concluded that the effects of the various compounds here described represent a true inhibition of D-amino-acid oxidase.

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Hydrolysis of Methylglucosides

In the course of a study of the mechanisms involved in the solvolytic reactions of glycosides, we have investigated the position of bond fission occurring in the hydrolysis of α- and β-D-methylglucosides. The reactions were carried out in water enriched in oxygen-18, and the methanol produced was isolated by fractional distillation in an efficient column, and then pyrolysed to give carbon monoxide which was examined by a mass spectrometer. We have found that the hydrolysis of α- and β-D-methylglucosides both in 1 *N* acid solution and in the presence of appropriate methylglucosidases proceeds by fission of the hexose–oxygen bond:

$$R|\text{—O—}CH_3 + H_2\,{}^{18}O \rightarrow R\text{—}{}^{18}O\text{—}H + HOCH_3$$

The results of typical experiments are shown in Table 1.

Table 1

Substrate	Catalyst	*p*H	Temp. (° C.)	Time of heating (hr.)	% excess abundance oxygen-18 in solution	% excess abundance oxygen-18 in methanol
α-D-Methylglucoside	1·10 *N* HCl	—	80	65	0·540	0·005
"	Yeast* α-methylglucosidase	6·5	35·7	6	0·300	0·000
β-D-Methylglucoside	1·10 *N* HCl	—	80	65	0·540	0·008
"	Almond emulsin	4·6	35·7	24	0·360	0·029
Blanks Glucose + methyl alcohol	1·10 *N* HCl	—	80	65	0·540	0·001
"	Yeast α-methylglucosidase	6·5	35·7	6	0·300	0·003
"	Almond emulsin	4·5	35·7	20	1·63	0·005

* A solution containing yeast α-methylglucosidase was obtained by killing, after washing and pressing, brewer's bottom yeast with liquid nitrogen and extracting the resultant autolysate with 0·1 *N* ammonia. Extracts prepared by autolysing the cells with toluene or ethyl acetate gave similar results.

Armstrong's[1] classical demonstration that α-D-glucose could be detected in the products of enzymolysis of α-D-methylglucoside, and that similarly β-D-glucose could arise from β-D-methylglucoside, has been used for relating the configurations of the glucosides to those of the parent sugars. The implicit assumption is, as Koshland[2] has pointed out, that no Walden inversion occurs. The present results show that since it is the hexose–oxygen bond which breaks, Armstrong's results cannot safely be used in this way. It can be said, however, that given the configurations on other grounds, Armstrong's work shows that under his conditions substitution at the $C_{(1)}$ atom occurs without a Walden inversion.

It is interesting to note that, both in the acid- and the enzyme-catalysed reactions, the position of bond fission is the same. The acid-catalysed reactions presumably proceed via the reversible formation of the conjugate acids of the glucoside followed either by rate-determining formation of a carbonium ion, with or without ring opening, or alternatively by bimolecular attack on the $C_{(1)}$ atom of the pyranose ring by a water molecule. The latter possibility, which seems unlikely on steric grounds, would, of course, involve a Walden inversion at the $C_{(1)}$ atom; but owing to the rapid mutarotation of the product under acid conditions, this test has no diagnostic value.

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