Table 5. COMPARISON OF THE ISOTOPIC TEMPERATURES AND DEPTH HABITATS OBTAINED FROM CORE *P*6304–8 (10 CM LEVEL) WITH THE TEMPERATURES AND DEPTH HABITATS OBSERVED BY JONES (1967) IN THE EQUATORIAL ATLANTIC

| Species | Temperature (°C) Isotopic *P*6304–8 (10 cm) | Temperature (°C) Observed Atlantic | Depth in water column (m) Calculated *P*6304–8 (10 cm) | Depth in water column (m) Observed Atlantic |
|---|---|---|---|---|
| *Globigerinoides ruber* | 29·1 | 14–29 | 20 | 0–100 |
| *G. trilobus trilobus* | 27·4 | 25–29 | 8–40 | 0–75 |
| *Orbulina universa* | 25·5 | 25–27 | 81 | 25–75 |
| *Pulleniatina obliquiloculata* | 25·2 | 25 | 85 | 25–50 |
| *Hastigerina pelagica* | 25·0 | 7–10 | 88 | 300–400 |
| *Globoquadrina dutertrei* | 22·2 | 24–27 | 121 | 25–50 |
| *Globorotalia truncatulinoides* | 18·0 | 14–17 | 181 | 100–200 |
| *G. crassaformis* | —— | 14–17 | —— | 75–200 |

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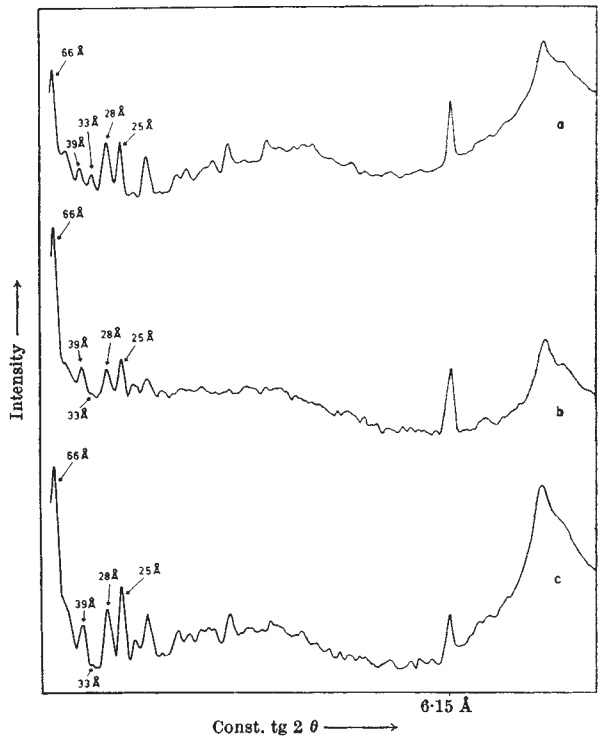


Fig. 1. Meridional photometer curves of the low angle X-ray diffraction patterns taken at room temperature with nickel-filtered CuK$\alpha$ radiation. *a*, Unstained mohair CSFH; *b*, mohair CSFH stained with 3,4,5-triiodobenzoic zcid *p*-nitrophenylester in DMF for 120 h at 50° C; *c*, mohair CSFH stained with *p*-(carboxymethylthiomercury)benzoic acid *p*-nitrophenylester in DMF for 24 h at 60° C.

## MOLECULAR STRUCTURE

# Further X-ray Evidence of Regularly Distributed Lysine in α-Keratin

IT has recently been shown that in mohair fibres the fifth order of the 198 Å pseudo repeat (39 Å reflexion) is intensified when the ε-amino groups of lysine residues are acylated with 3,4,5-triiodobenzoic acid *p*-nitrophenylester[1]. Particularly high yields of acylated ε-amino groups were obtained when dimethylsulphoxide was used as reaction medium. A sample of mohair which had been treated for 24 h at 40° C in this solvent, however, showed a somewhat diffuse X-ray pattern. We therefore looked for a reaction medium which did not cause structural distortion of the α-keratin and which gave high yields of acylated ε-amino groups.

Because dimethylformamide (DMF) is a suitable solvent for coupling reactions in peptide chemistry, mohair was treated with 3,4,5-triiodobenzoic acid *p*-nitrophenylester in DMF for 120 h at 50° C. In these conditions, 80 per cent of the ε-amino groups were acylated (DNP analysis). The meridional photometer curve of the X-ray pattern of this sample is shown in Fig. 1*b*. There is a relatively strong intensification of the 39 Å reflexion in comparison with the unstained sample (Fig. 1*a*).

In addition to the ester which was labelled with iodine we also used one which was labelled with mercury, as mercury has a smaller absorption factor and a higher atomic scattering power than iodine: *p*-(carboxymethylthiomercury)benzoic acid *p*-nitrophenylester was synthesized according to Matyash and Stepanov[2]. When the ε-amino groups were acylated with the mercury compound a partial decomposition of the mercury organic compound occurred.

The best method of acylation which we have found consists of treating mohair with *p*-(carboxymethylthiomercury)benzoic acid *p*-nitrophenylester in DMF for 24 h at 60° C. In these conditions only 65 per cent of the ε-amino groups are acylated (DNP analysis) but no decomposition is observed. In the X-ray pattern of such a sample (meridional photometer curve in Fig. 1*c*) there is also an intensification of the 39 Å reflexion, but the representation of this pattern in other reflexions is more comparable with that of the unstained sample. Molybdenum disulphide powder was used as an internal standard (6·15 Å reflexion).

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## CHEMISTRY

# Novel Facilitation of Peptide Synthesis

THE "solid phase" method of synthesizing peptides[1], in which the carbon-terminal residue is first esterified to a hydroxymethylpolystyrene resin, has been used successfully for the rapid synthesis in high yield of peptides which can be purified after removal from the resin[2]. If such a procedure, which omits the purification of intermediates, is to yield a homogeneous product without

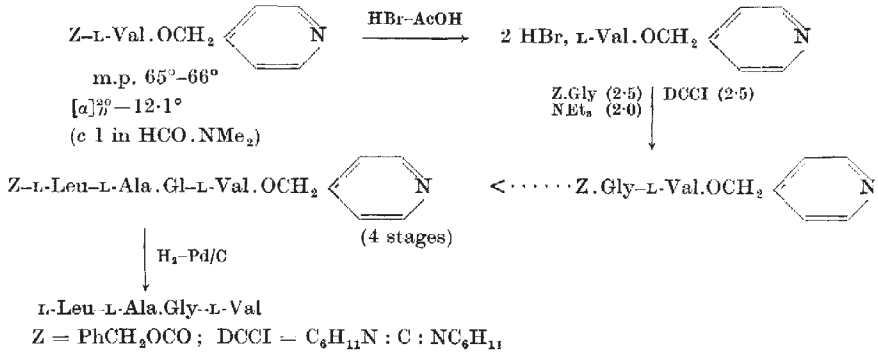


Fig. 1.

further purification, every stage must proceed to completion. For example, if 0·5 per cent of the amino-groups remain unacylated after each coupling, then after one hundred stages the product will contain only 61 per cent of the required peptide, the remainder being peptides with closely similar but defective sequences. It is a serious disadvantage of the solid phase method that one of the reactants is polymeric, making it difficult to force reactions to completion.

We are examining a fundamentally different procedure in which reactions are carried out in solution and the protected peptide is then adsorbed on an ion-exchanger while the excess of acylating agent and the co-products are washed away. For this purpose, a weakly basic group has been incorporated in the carbon-terminal residue by using its 4-picolyl ester (prepared from the reaction of the benzyloxycarbonylamino-acid triethylammonium salt with 4-picolyl chloride in dimethylformamide); these esters are stable to hydrogen bromide in acetic acid at room temperature but are cleaved by catalytic hydrogenation. The method[3] is illustrated by the synthesis of L-leucyl-L-alanyl-glycyl-L-valine, as outlined in Fig. 1. In the coupling reactions, 2·5 molar proportions of benzyloxycarbonylamino-acid and of dicyclohexylcarbodi-imide (relative to the amino-component) were used in tetrahydrofuran solution. After filtration to remove dicyclohexylurea, the coupling product was taken up on moist sulphoethyl 'Sephadex' (*C*-25, $H^+$ form), which was then washed thoroughly with tetrahydrofuran. The benzyloxycarbonylpeptide 4-picolyl ester was next eluted with 2 per cent triethylamine in aqueous tetrahydrofuran, and after evaporation of the solvent the benzyloxycarbonyl group was removed by treatment of the residue with 3·5 normal hydrogen bromide in acetic acid. The peptide ester dihydrobromide was precipitated with ether, and again from ethanol by ether, and the cycle was repeated. After the final hydrogenation in 80 per cent acetic acid, the solution was evaporated and the residue was dissolved in ethanol and precipitated by ether. Without further purification, the crude tetrapeptide monohydrate (60 per cent yield overall, from L-valine 4-picolyl ester dihydrobromide) was analytically and chromatographically pure; no alanylglycylvaline or glycylvaline could be detected by thin-layer chromatography which (in control experiments) could detect 0·2 per cent of these impurities. Amino-acid analysis of an acid hydrolysate gave the ratios: Leu, 0·96; Ala, 0·97; Gly, 1·00; Val, 1·00. A more sensitive test of the completion of each coupling stage was obtained by thin-layer chromatography of the intermediate benzyloxycarbonylpeptide 4-picolyl esters (a check not available in the solid-phase procedure); in every case, in conditions which will detect 0·1 per cent of the corresponding amino-component, none was found. The same tetrapeptide was obtained using *p*-nitrophenyl esters (4 molar proportions) for coupling (65 per cent overall yield of protected tetrapeptide). The specific rotations of the final products, $[\alpha]_D^{20} + 22·5°$ (*c* 1–2 in ethanol) (dicyclohexylcarbodi-imide method), $+23·7°$ (*p*-nitrophenyl ester method), are higher than those reported previously[1], $[\alpha]_D^{21} + 17·5°$, $+18·0°$, but the tetrapeptide (dicyclohexylcarbodi-imide method) was completely hydrolysed by leucineaminopeptidase (Leu, 1·00; Ala, 0·88; Gly, 0·98; Val, 1·04), and hydrogenation of the intermediate benzyloxycarbonylglycyl-L-valine 4-picolyl ester gave fully active glycyl-L-valine. A trace of a ninhydrin-positive contaminant, apparently arising from the 4-picolyl residues, always appeared in the products of hydrogenation of the picolyl esters; it is soluble in organic solvents and is therefore readily removed from normal peptides. This tetrapeptide is, however, unusually soluble in these solvents, and the contaminant was removed during drying at 100° C and 1 mm mercury. The protected tetrapeptide 4-picolyl ester was converted into the crystalline hydrazide by means of hydrazine hydrate, and this route should enable components which have been synthesized independently to be united through the acid azide. As a further variant, picolyl esters may be converted into amides in the usual way, and this should provide convenient syntheses of peptides having a carbon-terminal amide group. Such possibilities are now being examined.

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## Ultraviolet Photolysis of Sodium Nitrate Solutions in the Laboratory and by Sunlight

Chemical processes, induced by ultraviolet light in some aqueous inorganic nitrates, are of substantial interest in the chemistry of atmospheric aerosols. Nitrates form a large fraction of the inorganic natural aerosol[1] and, although they might be expected to decompose under sunlight[2], no systematic work on such photolysis has, as yet, been reported.

Earlier work on ultraviolet reduction of nitrate to nitrite was done by Villars[3] and Cultrera and Ferrari[4–6]. The effect was confirmed by others[7–10] and also in this laboratory, where it was found to be accompanied by the formation of highly oxidizing intermediates[11], and it was also found that the formation of nitrite is caused by primary steps in which higher oxides of nitrogen are involved[12–15]. The process of nitrite formation during photolysis of nitrates is reproducible as long as the solution is properly stirred[11], because stirring ensures the removal of gaseous reaction products from the zone below the interphase where