the usual isomerization of a 4-methoxyfuranoquinoline derivative into a N-methylfurano-4-quinolone compound (II).

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SYNTHESIS OF A SUBSTITUTED FRAGMENT A^{17-21} OF HUMAN INSULIN

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In connection with the development of a new scheme for the block synthesis of the A-chain of human insulin, we have performed the synthesis of the substituted pentapeptide (I) corresponding to fragment A^{17-21} of human insulin

$$\text{Bpoc-}\overset{17}{\text{Glu}}(\text{OBu}^t)\text{-Asn-Tyr}(\text{Bu}^t)\text{-Cys(Ec)-}\overset{21}{\text{Asn}}\text{-OBu}^t \qquad \text{(I)}$$

The pentapeptide (I) was synthesized by a 4 + 1 scheme using as starting materials the pentafluorophenyl ester of N-biphenylylisopropoxycarbonyl-γ-tert-butyl-L-glutamic acid (II), the pentafluorophenyl ester of N^{α}-benzyloxycarbonyl-L-asparagine (III), the methyl ester of N-benzyloxycarbonyl-O-tert-butyl-L-tyrosine (IV), S-ethylcarbamoyl-L-cysteine (V), and the tert-butyl ester of N^{α}-benzyloxycarbonyl-L-asparagine (VI).

Intermediate compounds were the methyl ester of O-tert-butyl-L-tyrosine (VII), the methyl ester of N^{α}-benzyloxycarbonyl-L-asparaginyl-O-tert-butyl-L-tyrosine (III), the methyl ester of L-asparaginyl-O-tert-butyl-L-tyrosine (IX), the methyl ester of N-biphenylylisopropoxycarbonyl-γ-tert-butyl-L-glutamyl-L-asparaginyl-O-tert-butyl-L-tyrosine (X), N-biphenylylisopropoxycarbonyl-γ-tert-butyl-L-glutamyl-L-asparaginyl-O-tert-butyl-L-tyrosine (XI), the pentafluorophenyl of N-biphenylylisopropoxycarbonyl-γ-tert-butyl-L-glutamyl-L-asparaginyl-O-tert-butyl-L-tyrosine (XII), N-biphenylylisopropoxycarbonyl-γ-tert-butyl-L-glutamyl-L-asparaginyl-O-tert-butyl-O-tyrosyl-S-ethylcarbamoyl-L-cysteine (XIII), the tert-butyl ester of L-asparagine (XIV), and the pentafluorophenyl ester of N-biphenylylisoproxycarbonyl-γ-tert-butyl-L-glutamyl-L-asparaginyl-O-tert-butyl-L-tyrosyl-S-ethylcarbamoyl-L-cysteine (XV).

For the synthesis of the S-ethylcarbamoyl-L-cysteine we used the method suggested by Guttmann [1]. The pentafluorophenyl esters of N-substituted amino acids and peptides were obtained by the method of Kisfaludy et al. [2].

The protected pentapeptide (I) was purified by recrystallization from dioxane—ethyl acetate (1:10). The structure of compound (I) was determined unambiguously by the scheme of synthesis, and its individuality was checked by chromatography and the results of analytical determinations.

tert-Butyl Ester of N-Biphenylylisopropoxycarbonyl-γ-tert-butyl-L-glutamyl-L-asparaginyl-O-tert-butyl-L-tyrosyl-S-ethylcarbamoyl-L-cysteinyl-L-asparagine (I). mp 192-193°C (decomp.), $[\alpha]_D^{23}$ − 38.6° (c, 1.0; dimethylformamide). TLC on Silufol UV-254 plates; R_f 0.30 (methanol—chloroform (1:9)), 0.52 (methanol—chloroform (2:17)). Found, %: C 60.49; H 7.05; N 9.87. $C_{53}H_{79}N_8O_{14}S$. Calculated, %: C 60.33; H 7.10; N 10.00. Amino acid analysis (the cysteine was not determined): Glu 1.00, Asp 1.96, Tyr 0.87.

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NOMOGRAM FOR THE RAPID DETERMINATION OF THE CONTRIBUTION OF SCATTERED LIGHT TO THE ABSORPTION SPECTRA OF BIOPOLYMER SOLUTIONS

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It is known that in the use of the absorption spectra of solutions of biopolymers (proteins, nucleic acids) in the ultraviolet region, in addition to the true absorption D_{true} due to the chromophores an additional optical density D_{sc} due to the Rayleigh scattering of the light is recorded, and

$$D_{true} = D_0 - D_{sc}, \quad (1)$$

where D_0 is the measured optical density.

It must be noted that D_{sc} may amount to 80% of D_0 [1], and therefore the measurement of D_{sc} is of prime importance.

At the present time it is customary to find D_{sc} by means of the relation

$$\lg D_{sc} = \lg a - n \lg \lambda, \quad (2)$$

which follows from the generalized formula of Rayleigh scattering [2]. However, it is extremely laborious and requires the expenditure of much time to find D_{sc} with the aid of formula (2).

The nomogram shown in Fig. 1 permits the process of finding D_{sc} to be considerably shortened and simplified. For this purpose it is sufficient to transfer the absorption spectrum pointwise to a nomogram, to find the region of wavelengths in which the points fall on a straight line (this region will correspond to the absence of true absorption), and to perform linear extrapolation. The value of D_{sc} must be found by recalculation from the extrapolation line with the ordinate corresponding to the selected wavelength. The resolving capacity of the nomogram is 0.2-0.3% of the optical density of the scattered light to be determined. The accuracy of measuring D_{sc} is determined by the accuracy of plotting the experimental points on the nomogram and the accuracy of extrapolation. The time necessary for determining D_{sc} is about 1 minute.

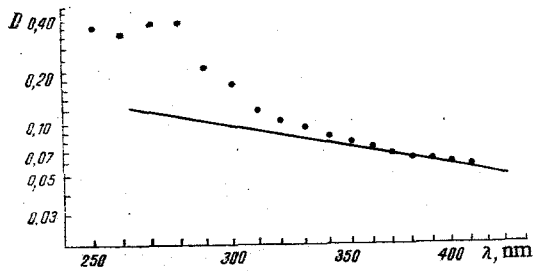

Fig. 1. Nomogram for determining the contribution of the apparent optical density due to the scattering of light in a measured absorption spectrum. The points show the absorption spectrum of an aqueous solution of thyroglobulin. The optical density of light scattering at 280 nm is 0.110.

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