14.5

II **

Table 1. Chemical composition of preparations of human thyroglobulin.

Preparation		Nitrogen content, % of dry weight	
I *	0.440	14.9	

* Preparation from thyroid tissue surgically removed from a case of nontoxic goiter.

0.324

** Preparation from normal thyroid glands obtained at autopsy.

Table 2. Ultracentrifugal analyses of preparations of human thyroglobulin.

Sedimentation const. Amounts of compo-(S_{20} , aq), of high molecular weight components in preparation const. Amounts of components, expressed as per cent of total high molecular weight material in prep.

Prep. I (cf. Table 1)	Prep. II	Prep. I	Prep. II
~ 7 S	6.9 S	3	3
11.6 "	11.7 "	11	6
17.8 "	17.1 "	81	86
23 "	26 "	5	5

values have been reported for thyroglobulin isolated from hog ^{6,7} and calf ⁵.

It therefore seems very probable that the main, high molecular weight component in the extracts studied by us represents human thyroglobulin.

The data given above suggest that the chemical and physico-chemical properties of human thyroglobulin are similar to those of thyroglobulin of animal origin.

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2,2'-Diphenyl-3,3'-diindolylmethane

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In a recent paper 1 on the chemistry of 3-hydroxymethylindoles we reported on their self-condensation to 3,3'-diindolylmethanes on boiling with water or aqueous sodium hydroxide. An exception was 3-hydroxymethyl-2-phenylindole which yielded 2-phenylindole on refluxing with 10 % sodium hydroxide. The expected product, 2,2'-diphenyl-3,3'-diindolylmethane (I), was described by Dahlbom and Misiorny who claimed to have obtained this compound from 2-phenylindole and formaldehyde. However, the melting point which

they reported (184—185°) for this substance seemed to be unusually low, since all the other known 3,3′-diindolylmethanes melt considerably above the corresponding indoles, as indicated in Table 1.

This led us to suggest that the compound, m.p. 184-185°, was not authentic 2,2'-

Table 1. Melting points of substituted indoles and 3,3'-diindolylmethanes. °C.

Substituent	Indole	3,3'-Diindolylmethane
None	52 - 53	167-168
1-Methyl	below 0	112.5 - 113
2-Methyl	62	237 - 238
1,2-Dimethyl	56	161.5 - 162.5
1-Methyl-2-		
phenyl	100 - 101	185 - 186
2-Phenyl	187 - 188	184 - 185

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diphenyl-3,3'-diindolylmethane. Dahlbom and Misiorny were quick to reply * to our unworthy suggestion, and in the light of further work to be described here, we offer our sincere apologies to these authors for suggesting that they had failed to perform a mixed melting point between 2-phenylindole and the product, m.p. 184—185°.

In the present work 2-phenylindole was refluxed in ethanol, containing a trace of hydrochloric acid, with ethyl orthoformate for 24 h. The product was 2,2'-diphenyl-3,3'-diindolylmethene which was isolated as its perchlorate (II) according to the procedure of Harley-Mason and Bu'Lock 4. Crystallization from acetic acid afforded orange-red prisms, m.p. 289-290° (decomp.) (Found: C 69.83; H 4.34; N 5.58. Calc. for C₂₉H₂₁O₄N₂Cl: C 70.09; H 4.26; N 5.64.) This methene salt was hydrogenated in ethanol in the presence of platinum oxide. Evaporation of the filtered solution yielded I which was obtained as colorless needles from ethanol, m.p. 188–189° (Found: C 87.50; H 5.77; N 6.78. Calc. for $C_{29}H_{22}N_2$: C 87.40; H 5.57; N 7.03). The di-1,3,5-trinitrobenzene complex of 2,2'diphenyl-3,3'-diindolylmethane was obtained as reddish brown needles from methanol, m.p. 161-162°(Found: C 59.86; H 3.62; N 13.82. Calc. for $C_{29}H_{22}N_2 \cdot 2C_6H_3O_6N_3$: C 59.71; H 3.42; N 13.59). In order to confirm that we had obtained authentic I, the compound m.p. 188-189° was Nmethylated according to the procedure of Potts and Saxton 5 to yield 1,1'-dimethyl-2,2'-diphenyl-3,3'-diindolylmethane, m.p. 185-186°, identical (mixed m.p., I.R. spectrum) with material previously obtained 1 by the self-condensation of 3-hydroxymethyl-1-methyl-2-phenylindole.

It is thus evident that Dahlbom and Misiorny obtained authentic I (the slight difference in our observed melting points for this compound is probably not significant) and it is remarkable that 2-phenylindole, 2,2'diphenyl-3,3'-diindolylmethane and its 1,1'-dimethyl derivative should melt so close together. We have also reinvestigated the self-condensation of 3-hydroxymethyl-2-phenylindole. In agreement with our previous results we obtained 2phenylindole when this compound was refluxed with 10 % sodium hydroxide. However, when exactly neutral or slightly acidic conditions were used the product was in fact the diindolylmethane I. We suspect that traces of alkali caused the formation of 2-phenylindole when 3-hydroxymethyl-2-phenylindole was refluxed with water in our previous experiments.

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Acid Soluble Nucleotide Linked Peptides in Extracts of Ascites Tumor Cells

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n a recent paper 1 it was reported that trichloroacetic acid (TCA) extracts of rabbit livers contained at least four ultra violet (UV) absorbing peaks with bound ninhydrin positive material. These four fractions were eluted immediately after adenosine diphosphate (ADP) and guanodiphosphate (GDP) with a formic acid-ammonium formate system² from a Dowex l column. The sitions on the elution curve of UV-linked ninhydrin positive material were in the same region where we previously had found the uridine nucleotide bound peptides in *L. casei* ³. Using a modification of the method of Strominger and Threnn ⁴ it was possible to isolate from the four fractions ten UV-absorbing spots. Seven of these contained from three up to seven amino acids, some of them at least bound to uridylic acid 5.

Since these data seemed to prove that animal tissues also contained acid soluble nucleotide bound peptides of a type previously only observed in extracts of different