compound is sensitive to moisture and oxygen, m.p. 37–38°, b.p. 105° (15 mm.). The infrared spectrum of this compound shows an absorption band at 12.8 μ. It is soluble in most organic solvents in all proportions, but reacts with alcohol forming an insoluble solid.

It is remarkable that the tetravalent chromium compound was isolated. Additional evidence for the tetravalent state of chromium was found in another experiment. An acid solution of potassium iodide was treated with a weighed amount of this compound and the liberated iodine titrated. Thus 0.638 g. of chromium tetra-*t*-butoxide liberated 1.89 mmol. of iodine. Assuming that one electron transfer occurred as shown in equation (1), the molecular weight was calculated as 338 in good agreement with the molecular weight determined cryoscopically.

$$Cr^{4+} + I^- \longrightarrow Cr^{3+} + {}^1/_2I_2 \qquad (1)$$

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GROSS STRUCTURE OF HEMOGLOBIN H

Sir:

Human hemoglobin H has been described in some detail by Rigas, Koler and Osgood.[1] Chemical investigations of chromatographically purified[2] hemoglobin H, here presented, lead to a further understanding of its structure and of its relation to other human hemoglobins.

When DNP-globin H was prepared and examined by methods previously described,[3,4,5] the result was approximately four N-terminal valyl residues per molecule of 66,000 molecular weight[1] but only one kind of N-terminal sequence: val-his-leu. This N-terminal sequence defines β chains[5] and suggests that hemoglobin H may be represented[6] as β_4^H.

"Fingerprints"[10] of tryptic hydrolysates of hemoglobins H and A differed markedly. Peptides numbered[10] 5, 10, 11, 13, 17, 18, 23, and probably several others in regions normally poorly resolved were absent on the fingerprint of H but no new peptides were apparent. The absent peptides were present on fingerprints of isolated α^A chains. The likely conclusion that the sequence in β^H and β^A chains is identical was substantiated by the following hybridization experiment.[8,11]

Following hybridization of carbonmonoxyhemoglobin H and radioactive carbonmonoxyhemoglobin S at *p*H 11.0 at 3° for 24 hr., four hemoglobins were chromatographically isolated. These data are pertinent:

	Reactants		Products			
Zone	...	...	1	2	3	4
Mg.	22	22	5[a]	2[a]	15	7
C.p.m./mg.	0	1200	70	1100	600	1200
Identity of material	Hb-H	Hb-S*	Hb-H	β_4^{S}*	Hb-A*	Hb-S*
Formula	β_4^A	$\alpha_2^A\beta_2^S$*	β_4^A	β_4^S*	$\alpha_2^A\beta_2^A$	$\alpha_2^A\beta_2^S$*

[a] Precipitation that occurred during hybridization must have consisted of β^A and β^S* chains because α chains are conserved.

Identification of the products involved chromatographic studies and determination of radioactivity and for hemoglobin A also the study of sedimentation velocity and examination of N-terminal peptides[3,4,5] to show that only the α chains were radioactive. Thus, hemoglobin A and β_4^S* were formed during hybridization but there was no evidence for $\beta_2^A\beta_2^S$*. On the basis of the radioactive and material balance, it was concluded that the four β chains of hemoglobin H are identical with each other and with β^A chains.

Hemoglobin H is the first observed example of a hemoglobin composed of a single kind of polypeptide chain. Possibly, other abnormal hemoglobins or minor components in normal hemoglobin may be built on the scheme α_4, $\alpha_3\beta$, $\beta_2\gamma_2$, etc. Biologically, it suggests that hemoglobin H disease results from an imbalance in the relative production of α and β chains and hence that α and β chains are under separate biosynthetic and genetic control. This latter suggestion is further supported by experiments now in progress which show that the α^A and α^F chains are identical and that β chains are present in several minor hemoglobin components normally associated with hemoglobin A and S.

These experiments were made possible by the interest and generosity of Dr. D. A. Rigas and Dr. R. D. Koler. This investigation was supported in part by grants H-2258 and H-3394 from the National Institutes of Health, United States Public Health Service.

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THE SYNTHESIS OF TIGOGENIN AND NEOTIGOGENIN

Sir:

We wish to report the synthesis of tigogenin (VIa) and neotigogenin (VIb), typical members of the large and important family of steroidal sapogenins.[1]

(1) *Cf.* L. F. Fieser and M. Fieser, "Natural Products Related to Phenanthrene," Reinhold Publishing Corp., New York, N. Y., 3rd Edition, 1949, Chapter VIII.

(1) D. A. Rigas, R. D. Koler and E. E. Osgood, *J. Lab. Clin. Med.*, **47**, 51 (1956).

(2) Extension of methods of D. W. Allen, W. A. Schroeder and J. Balog, THIS JOURNAL, **80**, 1628 (1958).

(3) H. S. Rhinesmith, W. A. Schroeder and L. Pauling, *ibid.*, **79**, 609 (1957).

(4) *Ibid.*, **79**, 4682 (1957).

(5) H. S. Rhinesmith, W. A. Schroeder and N. Martin, *ibid.*, **80**, 3358 (1958).

(6) The N-terminal sequence[5] defines the chain as α or β, the superscript denotes the hemoglobin that is the source of the chain, and the subscript has the usual chemical significance. The glycyl chains[7] of hemoglobin F are termed γ chains. Thus, hemoglobin A and S are $\alpha_2^A\beta_2^A$ and $\alpha_2^A\beta_2^S$ inasmuch as the α chains are identical.[8,9]

(7) W. A. Schroeder and G. Matsuda, THIS JOURNAL, **80**, 1521 (1958).

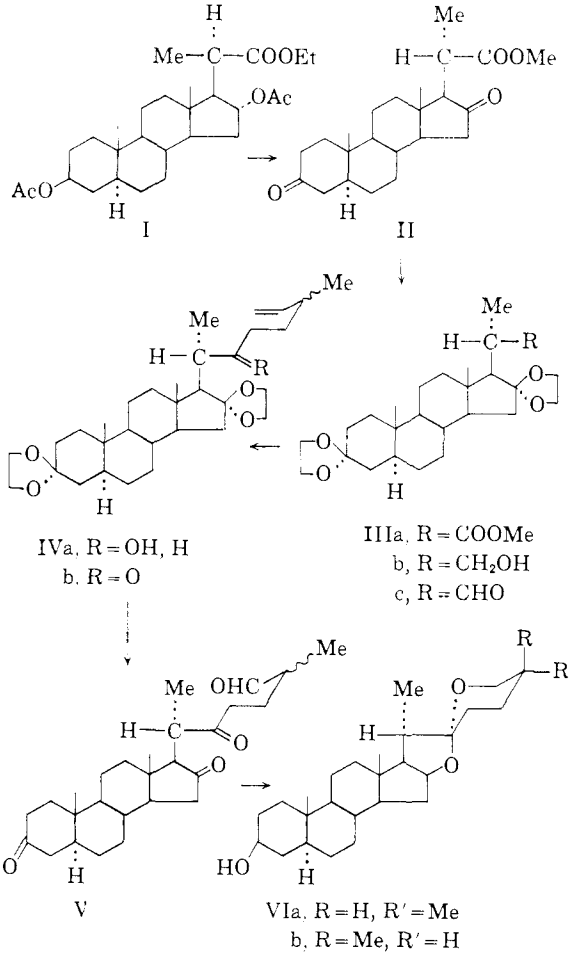
(8) J. R. Vinograd, W. D. Hutchinson, and W. A. Schroeder, *ibid.*, in press.

(9) V. M. Ingram, personal communication.

(10) V. M. Ingram, *Biochem. Biophys. Acta*, **28**, 539 (1958).

(11) J. Vinograd and W. D. Hutchinson, *Nature*, to be submitted.

We have described[2,3] recently the conversion by a three-step sequence of 3β,16α-diacetoxyandrostan-17-one to the 20-iso ethyl ester I,[4] which was transformed by three further steps to the 20-normal diketoester II.[3,5] The latter now has been converted to the corresponding di-cycloethylene ketal IIIa (60%) (m.p. 196–198°, $[\alpha]_D$ − 24°[6]).[7]



Lithium aluminum hydride (LAH) reduction yielded the primary alcohol IIIb (70%) (m.p. 235–237°, $[\alpha]_D$ − 18°),[8] which was oxidized with chromium trioxide–pyridine to the aldehyde IIIc (70%) (m.p. 183–184°, $[\alpha]_D$ − 21°).[9]

3-Methyl-4-pentenoic acid [b.p. 78–80° (25 mm.), $n^{20}D$ 1.4366][10] was reduced with LAH to 3-methyl-4-penten-1-ol [b.p. 63–64° (25 mm.), $n^{20}D$ 1.4369], which on treatment with phosphorus tribromide in pyridine yielded 1-bromo-3-methyl-4-pentene [b.p. 138–140° (764 mm.), $n^{20}D$ 1.4680]. The corresponding magnesium derivative on condensation with the aldehyde IIIc produced the alcohol IVa (70%) as a mixture of C-25 isomers (m.p. 143–157°, $[\alpha]_D$ − 18°),[11] which on oxidation with chromium trioxide–pyridine furnished the ketones IVb (85%) (m.p. 145–148°, $[\alpha]_D$ − 16°). Ozonolysis at − 18° in ethyl acetate–pyridine, and then decomposition with Raney nickel and treatment with dilute acetic acid, led to the triketo-aldehydes V which were reduced directly with sodium borohydride in isopropyl alcohol–tetrahydrofuran. Short heating of the product with dilute hydrochloric acid produced a 1:1 mixture of tigogenin (VIa) and neotigogenin (VIb) (25% from IVb.) Crystallization of the acetates[12] and saponification furnished neotigogenin (VIb) (m.p. 201–203°, $[\alpha]_D$ − 76°) identical with an authentic sample. Refluxing the synthetic mixture with ethanolic hydrochloric acid[13] for 120 hr. yielded tigogenin (VIa) (m.p. 202–204°, $[\alpha]_D$ − 68°), identical with an authentic specimen.

This work constitutes a total synthesis of the two steroidal sapogenins, since androstan-3β-ol-17-one is available by a number of total synthetic routes.[14] Further, in view of known interconversions, our work leads to the steroidal alkaloids of the tomatidine–solasodine type[15] as well as of the solanidine type.[16]

(2) N. Danieli, Y. Mazur and F. Sondheimer, *Chemistry & Industry*, 1724 (1958).

(3) N. Danieli, Ph.D. Thesis, Hebrew University, Jerusalem, June, 1958.

(4) The corresponding methyl ester now has been synthesized by a different route by V. Schwarz, V. Cerny and F. Sorm (*Chem. Listy*, **52**, 1633 (1958)).

(5) N. Danieli, Y. Mazur and F. Sondheimer, *Chemistry & Industry*, 1725 (1958).

(6) All rotations were measured in chloroform.

(7) J. W. Corcoran and H. Hirschmann, THIS JOURNAL, **78**, 2325 (1956).

(8) All new compounds gave satisfactory analytical results and the infrared spectra were compatible with the assigned structures. Yields are given to the nearest 5%.

(9) That no inversion at C-20 had occurred during the conversion of II to IIIc was shown through LAH reduction of IIIc to IIIb, which with dilute acetic acid and then with LAH yielded the same bisnorallocholane-3β,16β,22-triol (m.p. 247–250°, $[\alpha]_D$ +15°) as had been obtained[3] by the LAH reduction of II.

(10) *Cf.* W. J. Croxall and J. O. Van Hook, THIS JOURNAL, **72**, 803 (1950).

(11) The 22-hydroxyl group was apparently introduced stereospecifically, since the corresponding condensation of IIIc with isoamyl magnesium bromide yielded one single alcohol (m.p. 195–196°, $[\alpha]_D$ −20°).

(12) *Cf.* L. H. Goodson and C. R. Noller, *ibid.*, **61**, 2420 (1939); R. K. Callow and V. H. T. James, *J. Chem. Soc.*, 1671 (1955).

(13) *Cf.* R. B. Woodward, F. Sondheimer and Y. Mazur, THIS JOURNAL, **80**, 6693 (1958), and references cited there.

(14) *Cf.* J. W. Cornforth, "Progress in Organic Chemistry," Ed. J. W. Cook, Butterworths Scientific Publications, London, 1955, Vol. 3, Chapter 1.

(15) (a) F. C. Uhle, THIS JOURNAL, **76**, 4245 (1954); (b) F. C. Uhle and J. A. Moore, *ibid.*, **76**, 6412 (1954).

(16) Y. Sato and H. G. Latham, *ibid.*, **78**, 3146 (1956); see also F. C. Uhle and W. A. Jacobs, *J. Biol. Chem.*, **160**, 243 (1945).

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1,2-DIFERROCENYLETHANE FROM AN UNUSUAL REACTION

Sir:

Ferrocene condenses with formaldehyde in either concentrated sulfuric acid[1] or liquid hydrofluoric acid[2] to give a compound (I) containing two ferrocene nuclei and two methylene groups (from two moles of formaldehyde). This condensation product (I) was originally assigned the structure 1,1′-bis-ferrocenylenemethane (Ia).[1,2,3] Subsequently, Nesmeyanov and co-workers[4] revised their assign-

(1) A. N. Nesmeyanov and I. I. Kritskaya, *Izvest. Akad. Nauk S.S.S.R., Otdel. Khim. Nauk*, 253 (1956).

(2) V. Weinmayr, THIS JOURNAL, **77**, 3009 (1955).

(3) Y. T. Struchkov, *Zhur. Obshchei Khim.*, **27**, 2039 (1957).

(4) A. N. Nesmeyanov, L. A. Kazitsyna, B. V. Lokshin and I. I. Kritskaya, *Doklady Akad. Nauk S.S.S.R.*, **117**, 433 (1957).