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Electroorganic Preparations 1. Reduction of Steroid Ketones

HENNING LUND

Leo Pharmaceutical Products, Copenhagen, Denmark

It is shown that Δ^4-keto-3-steroids and $\Delta^{1,4}$-keto-3-steroids can be reduced at a mercury cathode at controlled cathode potential to pinacols, whereas saturated keto groups are not reduced at the potential used. The stereochemistry of the pinacols is discussed. The formation and a subsequent oxidation of the pinacols with a 1,2-glycol splitting reagent as lead tetraacetate or periodic acid is proposed as a method for protection of the reactive unsaturated keto group in the presence of saturated keto groups.

Δ^4-Unsaturated steroid ketones are known to be reducible at the dropping mercury electrode [1–4], but no conclusive evidence of the electrode reaction has been presented. The purpose of the present work is to examine the electrode reaction and its use in preparative chemistry. The investigation consists of a polarographic examination of the compounds under different conditions and controlled potential electrolysis under similar conditions.

POLAROGRAPHIC INVESTIGATIONS

The half-wave potentials of progesterone and androsta-1,4-diene-17β-ol-3-one in 75 % methanol—25 % water containing 0.5 M LiCl and 0.05 M buffer are listed in Table 1. The compounds showed only one wave when the solutions had been deaerated for half an hour. The slope of $E_{\frac{1}{2}}$ *vs* pH is near the value 0.059 required for reversible reactions [5].

The height of the wave varies only slightly with pH indicating that the same number of electrons participate in the electrode reaction in acid and alkaline solution. The reduction of an $\alpha,\beta,\alpha',\beta'$-unsaturated ketone must take place at the carbonyl group as the reduction of a double bond would yield an α,β-unsaturated ketone that would show a reduction wave.

The diffusion coefficients D are calculated from the diffusion currents by means of the Ilkovic equation [5]. As the product of D and the molecular weight M is the same for the two compounds it is likely that they use the same number of electrons in the electrode reaction.

The pH-values are measured with a glass electrode, but it was felt necessary to control the found values in the medium used by measuring the half-wave potentials of anthraquinone-2-sulfonate in the same solutions. By assuming a linear connection between $E_{\frac{1}{2}}$ for anthraquinone-2-sulfonate and pH, the pH of the solution used was found as the abscissa-value corresponding to the measured $E_{\frac{1}{2}}$-ordinate. The two methods gave identical results from pH 1 to pH 11.

The polarographic results can be accounted for by the following assumptions. The potential determining step of the electrode reaction is reversible in the examined pH-interval. The number of electrons in the electrode reactions for α,β-unsaturated and $\alpha,\beta,\alpha',\beta'$-unsaturated steroid ketones is the same and probably one from pH 1 to 11. The similarity of the polarographic behaviour of an α,β- and an $\alpha,\beta,\alpha',\beta'$-unsaturated steroid ketone makes it likely that an α,β-unsaturated steroid ketone also is reduced at the carbonyl group. The polarographic behaviour of an α,β-unsaturated steroid ketone is thus different from that of the simple α,β-unsaturated ketones [6].

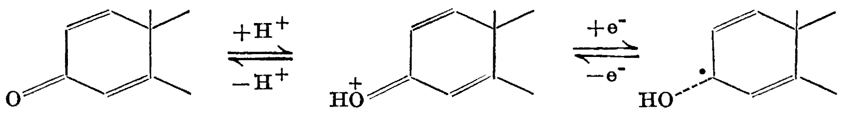
Table 1. **Half-wave potential (*vs.* saturated silver/silver chloride electrode) and diffusion current at different pH's for progesterone and androsta-1,4-diene-17β-ol-3-one. Drop time *t* measured at $E_{\frac{1}{2}}$.**

	Progesterone			Androstadieneolone		
Concentration	1.85×10^{-3} M			2.32×10^{-3} M		
D	7.0×10^{-6} cm²/sec			7.6×10^{-6} cm²/sec		
$D \times M$	2.18×10^{-3}			2.18×10^{-3}		
pH	$-E_{\frac{1}{2}}$ V	i_d μA	t sec	$-E_{\frac{1}{2}}$ V	i_d μA	t sec
1.05	1.01_3	5.00	3.08	0.87_5	6.60	3.15
4.00	1.18	4.95	3.05	1.06	6.60	3.14
5.08	1.24_7	4.94	3.00	1.11_3	6.45	3.11
9.80	1.56_2	4.75	2.70	1.41_2	6.25	2.84
10.16	1.58_3	4.75	2.62	1.42_5	6.25	2.83
11.0	1.67	4.70	2.43	1.48_8	6.20	2.73

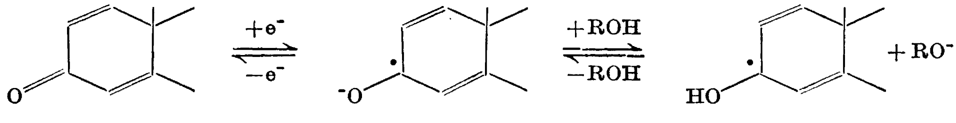
PREPARATIVE REDUCTIONS

The controlled potential electrolysis showed that α,β-unsaturated and $\alpha,\beta,\alpha',\beta'$-unsaturated steroid ketones yielded pinacols both in acid and alkaline solution and are thus different from the simple α,β-unsaturated ketones which dimerise at the β-carbon atom [6]. The reaction path of the reduction of an $\alpha,\beta,\alpha',\beta'$-unsaturated steroid ketone may be formulated as follows:

Acid solution:



Alkaline solution:



The radical formed dimerises slowly and irreversibly thus forming a pinacol. The products were proved to be pinacols by their ability to form the original ketones on oxidation with lead tetraacetate. The dimerisation takes place at carbon atom number 3 rather than at 1 or 5, as a dimerisation at one of the latter carbon atoms would be subject to severe steric hindrance.

The polarographic reduction is not the only reduction in which the α,β-unsaturated steroid ketones behave in another way than the simple unsaturated ketones. Windaus [7] found that cholestenone yielded a pinacol by reduction with sodium amalgam, and as he expected to get a saturated dimerised ketone from an α,β-unsaturated ketone he concluded that cholestenone was not an α,β-unsaturated ketone.

The reduction of a Δ^4-3-keto-steroid to a pinacol gives rise to the formation of two asymmetric carbon atoms and there is thus the possibility of formation of three compounds, one with both hydroxyl groups α, one with both hydroxyl groups β and one with one α and one β hydroxyl group.

An investigation of a model shows that in the compound where both hydroxyl groups are α the ring A will be in the 'half-chair' conformation and the bonding between the two dimerised radicals 'quasi-equatorial'. When the hydroxyl groups are β it is likely that the ring A adopts the 'half-boat' conformation and the hydroxyl groups are thus 'quasi-flagpole' and the carbon-carbon bond 'quasi-bowsprit'. In that case there will be free rotation of the carbon-carbon bond and the hydroxyl groups would then have the possibility of being *cis* which would facilitate the oxidation of the glycol with lead tetraacetate.

The relative amounts of the possible isomers depend among other things upon pH. Thus androsta-1,4-diene-17β-ol-3-one yields at pH 5 almost exclusively a pinacol with the rotation $[\alpha]_D^{23} = +277°$, whereas it in alkaline solution nearly quantitatively yields a pinacol with the rotation $[\alpha]_D^{23} = +8°$. The pinacol formed in acid solution dehydrates very easily by treatment with acid, whereas the one formed in alkaline solution requires more drastic conditions for the dehydration.

It is tempting to explain this by assuming that in acid solution it is the protonated ketone which is reduced, whereas the free ketone is reduced in alkaline solution. The most likely way for a steroid molecule to get the conjugated ketone system in contact with the cathode is by approaching the cathode with the methyl groups in the positions 10 and 13 pointing away from the electrode. In acid solution the positive protonated carbonyl oxygen would be attracted by the negative cathode, and the formed pinacol would thus have the hydroxyl group α. In alkaline solution the negative carbonyl oxygen would be repulsed from the cathode to the β configuration, where it also has the best chance of extracting a proton from a solvent molecule.

The proposed configurations are consistent with the difference found in the rate of dehydration. It has been shown [8] that divinyl carbinols very easily by an allylic rearrangement form a carbinol with the double bonds conjugated. According to Stork and White [9] there is a *cis* relation between the departing and the entering group in a S_N2' allylic rearrangement. A S_N1' mechanism must be rejected as it does not distinguish between the two isomers. The α-hydroxyl pinacol would thus yield a 5-α-carbinol, where the hydroxyl group is axial. As the 6-β-hydrogen is axial too, the 5-α-carbinol would dehydrate smoothly on treatment with acid. The β-hydroxyl pinacol would not have the same chance for dehydration as there is less tendency to form a 5-β-carbinol and no axial α-hydrogen in position 6 or 10.

It has furthermore been pointed out [10] that there is a difference in rotation from 200° to 300° between the α-hydroxy and the β-hydroxy isomer of unsaturated 3-hydroxy steroids as Δ^4-cholestenol ($\alpha - \beta = +297°$) and cholesta-4,7-dien-3-ol ($\alpha - \beta = +269°$). The difference in rotation between the proposed α-hydroxyl pinacol and the β-hydroxyl pinacol is $+269°$.

For the Δ^4-steroid ketones the difference in the composition of the mixture of pinacol isomers formed in acid and alkaline solution is not as pronounced as in the case of $\Delta^{1,4}$-steroid ketones. This is probably due to the fact that whereas in the latter the ring A is planar or nearly planar the former has the possibility of existing in a 'half-chair' or a 'half-boat' conformation. Etiocholane-3,17-dione exists partly with ring A in the boat form [11] and if it is assumed that the same is the case for the unsaturated 3-ketones, this explains the formation of a mixture of the pinacol isomers, as the reduction of the 'half-chair' and the 'half-boat' may result in the formation of different isomers. The isomer with the hydroxyl groups in the β-configuration probably has the ring A in the 'half-boat' conformation and it may, therefore, be impossible to deduce anything concerning the configuration from the rotations.

Like other 1,2-glycols the pinacols are quantitatively oxidized at room temperature by lead tetraacetate to carbonyl compounds the carbon-carbon bond being cleaved, thus forming the original α,β-unsaturated ketones. This reaction has been used to prove the reduction products to be 1,2-glycols.

The reduction of an α,β-unsaturated keto group to a pinacol and the subsequent oxidation of the pinacol to an α,β-unsaturated carbonyl group may be used to protect the more reactive conjugated carbonyl group, while the less reactive saturated keto group is brought to reaction with some other reagent, which does not attack the pinacol system. After the reactions the pinacol is oxidized with lead tetraacetate or periodic acid to the α,β-unsaturated carbonyl group.

The pinacols are slowly, but well crystallizing compounds which mostly are less soluble than the ketones from which they are formed. Sometimes a pinacol crystallizes in two forms with different solubilities and it is not always easy to get the more soluble form to crystallize in the less soluble form [12]. The solubilities of the isomers are qualitatively the same but quantitatively there may be quite a difference. They are all soluble in tetrahydrofuran, soluble or recrystallizable from amyl acetate, toluene and ethyl alcohol and slightly soluble in water and hexane. The pinacols are stable in alkaline solution, whereas in acid solution they more or less rapidly lose water thus forming a compound with four (for the diene six) conjugated double bonds [13]. Dilute acetic acid affects the pinacols very slowly, whereas hydrochloric acid acts very quickly as dehydrating agent. Even the small amount of hydrochloric acid present in ordinary chloroform makes it impossible to use chloroform as a solvent for the pinacols for more than a short period. That is the reason why tetrahydrofuran has been used as solvent in the measurements of the rotation of the compounds. The pinacols themselves are also dehydrated at elevated temperatures, a fact which makes the melting point of minor importance as a physical constant of the compound.

EXPERIMENTAL

Apparatus. To the polarographic investigation were used a Sargent-Heyrovsky Model XII photographically recording polarograph and a recording polarograph Radiometer Type PO 3 a; the reported results are obtained with the latter. The capillary delivered 2.346 mg of mercury per second at an effective column height of 48 cm. The drop times reported were measured in the test solutions at the half-wave potential of the reducible compound.

All potentials are measured against a silver/silver chloride electrode in an aqueous saturated potassium chloride solution separated from the test solution with an agar plug and fritted glass diaphragm.

The potentiostat was a Lingane-Jones potentiostat [14]. For manual control a circuit adapted from Lingane [15] was used, in which the D.C. source was a 12 volts accumulator, the voltage adjuster a variable resistance (50 Ω) and the potential measuring device a "Polymeter" manufactured by Radiometer, Copenhagen, which had a resistance of 12×10^6 Ω.

The cell for the preparative reduction is adapted from Pasternak [6]. It consists of two conical 200 ml flasks which are connected through two fritted glass diaphragms 30 mm in diameter. In the cathode chamber is placed the mercury cathode (34 cm^2) which is connected through a platinum wire through the bottom of the flask to the outer circuit, the silver/silver chloride reference electrode, which has the fritted glass diaphragm just above the cathode, and a dropping mercury electrode. Further there are a stirrer (a uniform stirring is important) and an inlet for purified [16] nitrogen. Between the diaphragms agar containing potassium chloride or some other suitable salt is placed, and in the anode chamber is the platinum anode (15 cm^2). When the cell contains a 2 M salt solution the resistance is about 10 Ω.

Materials. The buffer solutions were made according to Ref.[17]. The materials were analytical grade. The steroids were recrystallized to constant properties.

Procedure. The controlled-potential electrolysis were performed in the following way. In the cathode chamber is placed 150 ml of the reaction medium (aqueous buffer solution and methanolic lithium chloride solution) and the anode chamber is filled with aqueous, alkaline sodium nitrite. The cathode chamber is deaerated with purified nitrogen [16] and reducible impurities are removed by electrolysis by keeping the cathode potential 0.3 – 0.4 volts more negative than the half-wave potential of the compound to be reduced. When the current has reached a stationary value, the reducible compound is added. By use of a potentiostat or by manual control the cathode potential is kept 0.1 – 0.2 volt more negative than the half-wave potential of the compound in the medium used. The current drops from an initial value of 0.5—0.3 A to about 5 mA during 3—4 h. The reduction is followed polarographically with the dropping mercury electrode in the reaction medium whereby it is ensured that the reduction is undertaken at the right potential and the progress of the reduction can be followed. When the polarogram shows that the reduction is complete, the process is discontinued.

ISOLATION OF THE PINACOLS

The cathode chamber is emptied, the reaction medium is diluted with 200 ml water and made alkaline, if that does not affect the side chain of the steroid. The pinacols are extracted with a mixture of tetrahydrofuran and ether. After washing the organic layer with a bicarbonate solution and water, the solvents are evaporated at temperatures not exceeding 40°.

Androstenedione pinacol (1). 1.00 g of androstenedione was reduced at pH = 5 at —1.30 V. The pinacol residue was refluxed half an hour with 20 ml ethanol until a uniform suspension was obtained. After cooling for two days 690 mg (compound A) were filtered off, which after recrystallization from 25 ml toluene yielded 600 mg. $[\alpha]_D^{23} = + 153° \pm 2°$ (tetrahydrofuran, $c = 0.438$, $a = 1.34°$, $l = 2$ dm). The compound dehydrates before melting. The dehydrated compound decomposes at about 305°. (Found: C 79.45; H 9.45. Calc. for $C_{38}H_{54}O_4$: C 79.47; H 9.47). The infra-red and ultra-violet spectrum and polarographic data show that no conjugated carbonyl group is present and the ability to form a semicarbazone and the infra-red spectrum show the presence of a keto group. The compound yields a yellow compound on treatment with acid and it yields quantitatively androstenedione by oxidation with lead tetraacetate in dry benzene at room temperature. From these pieces of evidence it is concluded that the compound A is one of the possible isomers of androstenedione pinacol.

The mother liquor was evaporated and the residue recrystallized from 3 ml ethanol. There was obtained 80 mg of compound A identified by its rotation and its ability to form androstenedione by oxidation with lead tetraacetate.

Androstenedione pinacol (2). 1.00 g of androstenedione was reduced at pH about 12.5 at —1.70 V. The pinacol residue was boiled with 20 ml 96 % ethanol until a uniform suspension was obtained. After cooling for two days 420 mg was filtered off, which after recrystallization from 15 ml toluene (360 mg) had a rotation of $[\alpha]_D^{23} = +152° \pm 2°$ (tetrahydrofuran, $c = 0.558$, $a = 1.70°$, $l = 2$ dm) (Found: C 79.43; H 9.40. Calc. for $C_{38}H_{54}O_4$: C 79.47; H 9.47). As all the chemical and physical data are indistinguishable from those of androstenedione pinacol A it was concluded that the isolated compound was the same isomer of androstenedione pinacol. The mother liquor was evaporated and the residue dissolved in benzene. On addition of hexane a component is obtained (410 mg) which after recrystallization from alcohol-water (340 mg) has a rotation of $[\alpha]_D^{23} = +174° \pm 2°$ (tetrahydrofuran, $c = 0.705$, $a = 2.45°$, $l = 2$ dm). The compound shrinks about 160° and dehydrates. The dehydrated compound decomposes at about 300°. (Found: C 79.45; H 9.48. Calc. for $C_{38}H_{54}O_4$: C 79.47; H 9.47). This compound had an infra-red spectrum very similar to that of compound A, and as it yielded androstenedione by oxidation with lead tetraacetate it was concluded, that this compound B was another isomer of androstenedione pinacol.

Progesterone pinacol (1). 1.00 g of progesterone was reduced at pH = 5 at —1.30 V. The pinacol residue was boiled with 20 ml ethanol until a uniform suspension was obtained. After cooling for two days 650 mg (compound PA) were filtered off. It was recrystallized from 20 ml acid-free amyl acetate. Rotation $[\alpha]_D^{23} = +162° \pm 2°$ (tetrahydrofuran, $c = 1.27$, $a = 4.10°$, $l = 2$ dm). The compound dehydrates before melting. The dehydrated product decomposes at about 290°. (Found: C 79.68; H 10.05. Calc. for $C_{42}H_{62}O_4$: C 79.94; H 9.91). The compound is soluble in tetrahydrofuran, less soluble in ethanol and benzene and insoluble in water and hexane. The infra-red spectrum shows the presence of an unconjugated carbonyl group and the absence of a conjugated ketone. This is confirmed by polarographic data and ability to form a semicarbazone. The compound PA yields a yellow compound on treatment with acid or heating to about 190°. It gives by oxidation with lead tetraacetate at room temperature in dry benzene quantitatively progesterone. It is, therefore, concluded that the compound PA is one of the possible isomers of progesterone pinacol.

The mother liquor was evaporated and the residue dissolved in 4 ml ethanol. On standing was obtained 65 mg of a compound which was identified as compound PA by its rotation and ability to form progesterone by oxidation with lead tetraacetate.

Progesterone pinacol (2). 1.00 g of progesterone was reduced at pH 12.5 at −1.70 V. The pinacol residue was refluxed with 20 ml ethanol for half an hour. After cooling for two days a compound (560 mg) was filtered off. The rotation was not changed by recrystallization from amyl acetate. $[\alpha]_{\mathrm{D}}^{23} = +163° \pm 2°$ (tetrahydrofuran, $c = 0.675$, $a = 2.20°$, $l = 2$ dm). M. p. (after dehydration) 285−290° (decomp.) (Found: C 79.80; H 9.79. Calc. for $C_{42}H_{62}O_4$: C 79.94; H 9.91). As all the physical and chemical data for the compound are indistinguishable from those of progesterone pinacol PA it was concluded that the isolated compound was the same isomer of progesterone pinacol.

The filtrate from compound PA was evaporated and the residue dissolved in hot benzene. On addition of hexane a compound is obtained (280 mg) which after recrystallization from ethanol-water yields 190 mg. $[\alpha]_{\mathrm{D}}^{23} = +186° \pm 2°$ (tetrahydrofuran, $c = 0.536$, $a = 1.99°$, $l = 2$ dm). M. p. 165°. On further heating it dehydrates and solidifies at about 180° and the dehydrated compound decomposes at about 285°. (Found: C 79.43; H 9.84. Calc. for $C_{42}H_{62}O_4$: C 79.94; H 9.91). The compound has an infra-red spectrum very similar to that of compound PA and as it yielded progesterone by oxidation with lead tetraacetate it was concluded that the compound PB was an isomer of progesterone pinacol.

Androstadieneolone pinacol (1). 1.00 g of androsta-1,4-diene-17β-ol-3-one was reduced at pH = 5 at −1.20 V. The pinacol residue was dissolved in benzene. On addition of hexane a compound is obtained (850 mg) with a rotation of $[\alpha]_{\mathrm{D}}^{23} = +277° \pm 2°$ (tetrahydrofuran, $c = 0.542$, $a = 3.11°$, $l = 2$ dm). M. p. after dehydration 208−211° (decomp.). (Found: C 79.10; H 9.49. Calc. for $C_{38}H_{54}O_4$: C 79.47; H 9.47). The compound is very easily dehydrated under influence of acid, light or heat and yields a yellow compound. The infra-red spectrum shows absence of a carbonyl group. By oxidation with lead tetraacetate in dry benzene androstadieneolone is the main product, but a minor part of the compound is dehydrated by the acetic acid. All the data are consistent with the assumption that the isolated compound is one of the isomers of androstadieneolone pinacol.

The mother liquor was evaporated and evidence of a compound with lower rotation was found, but the compound was not isolated.

Androstadieneolone pinacol (2). 0.620 g of androstadieneolone was reduced at pH = 12.5 at −1.60 V. The pinacol residue was recrystallized from benzene yielding 510 mg of a compound with a rotation of $[\alpha]_{\mathrm{D}}^{23} = +8° \pm 2°$ (tetrahydrofuran, $c = 0.683$, $a = 0.11°$, $l = 2$ dm). M. p. 180−182° (decomp.). (Found: C 79.63; H 9.30. Calc. for $C_{38}H_{54}O_4$: C 79.47; H 9.47). This compound is not dehydrated as easily as the above mentioned isomer of androstadieneolone pinacol, but can be dehydrated by heating to about 200° or by treatment with hydrochloric acid. It is quantitatively oxidized by lead tetraacetate at room temperature to androstadieneolone. It contains no carbonyl absorption in the infra-red spectrum. From the evidence it is concluded that the compound is a second isomer of androstadieneolone pinacol.

The mother liquor was evaporated and evidence of a compound with higher rotation was found, but the compound was not isolated.

Cholestenone pinacol. 1.00 g of cholestenone was reduced at pH = 5 at −1.30 V. The pinacol residue was refluxed with 25 ml ethanol for two hours. After cooling a compound (850 mg) was obtained, which after recrystallization from amyl acetate has a rotation of $[\alpha]_{\mathrm{D}}^{23} = +92° \pm 2°$ (tetrahydrofuran, $c = 3.052$, $a = 5.65°$, $l = 2$ dm). Dolou *et al.*[13] report $[\alpha]_{\mathrm{D}}^{23} = 85°$, but a recalculation of their data gives 92°. (Found: C 84.12; H 11.72. Calc. for $C_{54}H_{90}O_2$: C 84.08; H 11.77). The compound has physical and chemical data consistent with the data reported [12,13] for cholestenone pinacol. The compound yields quantitatively cholestenone by oxidation with lead tetraacetate.

Testosterone propionate pinacol. 1.00 g of testosterone propionate was reduced at pH = 5 at −1.30 V. The pinacol residue was recrystallized from ethanol giving 700 mg of a compound with a rotation of $[\alpha]_{\mathrm{D}}^{23} = +76° \pm 2°$ (tetrahydrofuran, $c = 0.586$,

$a = 0.90°$, $l = 2$ dm). (Found: C 76.68; H 9.46. Calc. for $C_{44}H_{66}O_6$: C 76.46; H 9.63). The found data are consistent with the data reported [12] for testosterone propionate pinacol. It possesses no androgenic activity.

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REFERENCES

1. Eisenbrand, J. H. and Picher, H. *Hoppe-Seylers Z. Physiol. Chem.* **260** (1939) 83.
2. Wolfe, J. K., Hershberg, F. B. and Fieser, L. F. *J. Biol. Chem.* **136** (1940) 653.
3. Zuman, P., Tenygl, J. and Brezina, M. *Collection Czechslov. Chem. Communs.* **19** (1954) 46.
4. Kabasakalian, P. and McGlotten, J. *J. Am. Chem. Soc.* **78** (1956) 5032.
5. Kolthoff, I. M. and Lingane, J. J. *Polarography*, Interscience Publishers, New York, London 1952.
6. Pasternak, R. *Helv. Chim. Acta* **31** (1948) 753.
7. Windaus, A. *Ber.* **39** (1906) 518.
8. Heilbron, I. M., Jones, E. R. H., McCombie, J. T. and Weedon, B. C. L. *J. Chem. Soc.* **1945** 85.
9. Stork, G. and White, W. N. *J. Am. Chem. Soc.* **75** (1953) 4119.
10. Mills, J. A. *J. Chem. Soc.* **1952** 4976.
11. Nace, H. R. and Turner, R. B. *J. Am. Chem. Soc.* **75** (1953) 4063.
12. Butenandt, A. and Poschmann, L. *Ber.* **73** (1940) 893.
13. Dolou, R., Chopin, J. and Raoul, Y. *Bull. soc. chim.* **18** (1951) 646.
14. Lingane, J. J. and Jones, S. L. *Anal. Chem.* **22** (1950) 1169.
15. Lingane, J. J. *Electroanalytical Chemistry*, Interscience Publishers, New York, London 1953, p. 202.
16. Meites, L. and Meites, T. *Anal. Chem.* **20** (1948) 984.
17. *Pharmacopoea Danica* 1948 I, p. 222.

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