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Improved Microcombustion Technique for Carbon-Hydrogen Estimation in Perhydro-polycyclic Compounds. I

Steroids and Triterpenoids*

By

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Introduction

Polycyclic compounds are known to give low carbon values by the conventional microanalytical method[1]. During routine analysis over the last 15 years it has been found that perhydropolycyclic compounds, particularly those having angular methyl groups, give erroneous results using copper oxide or copper oxide-lead chromate filling.

Analytical data of steroids, triterpenoids and similar compounds of natural origin reported in the literature frequently show more than 0.5 per cent deviation in carbon values[2–11]. In general the values are less for carbon than the calculated ones. It was, therefore, of interest to improve upon the existing methods and the present communication deals with such an attempt.

Experimental

Equipment and Reagents

Microcombustion tube, Vycor, with side arm, from Arthur H. Thomas Co., Philadelphia, U. S. A.

Cupric oxide, wire form, A. C. S. specifications, "Baker Analyzed" Reagent.

Asbestos, platinised 30%, Arthur H. Thomas Co.

Silver metal wool for microchemical procedures, Arthur H. Thomas Co.

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Lead dioxide, C. P., from Coleman & Bell Co., Norwood, U. S. A.

Platinum tetrahedra, Arthur H. Thomas Co., Catalogue No. 8313 (1965), p. 866.

Procedure

Procedure A. The combustions were carried out on a microcombustion assembly for carbon-hydrogen estimation [Arthur H. Thomas Co., Philadelphia, U. S. A., Catalogue No. 6447-C (1965), p. 693]. The "Combination filling" consisted of a silver plug at the capillary end followed by a lead dioxide layer 3 cm long, a 4.5 cm layer of silver metal wool, a 11.5 cm layer of copper oxide and a 3-cm roll of platinum gauze (the different layers were separated from each other by a pad of 1 mm thick platinized asbestos) and the combustion furnace and kept at $700 \pm 25°$. The rate of flow of oxygen was regulated at 5 ml per minute and the microboat was kept at a distance of 5 cm from the stationary furnace. The temperature of combustion in the electric sample heater was $900 \pm 25°$. The analysis was carried out as described by *Niederl* and *Niederl*[12].

Procedure B. The "simple band filling" was used; it consisted of three layers of copper oxide alternated with two layers of platinized asbestos each 2.5 cm long, followed by a 0.5-cm layer of silver wool and a 2-cm layer of platinum tetrahedra preceding the bands. Heraeus' Microcombustion Furnace (Type F, No. 3–14) was used in which it was possible to attain a temperature upto 1200°. The temperature of the stationary furnace (Langbrenner) was maintained at $900 \pm 25°$ and the rate of flow of oxygen was regulated at 4 to 5 ml per minute. The sample heater (Kurzbrenner) was driven at 15 mm per minute and its temperature was kept over 1000° by adjusting the needle of the "Indicator Gauge" at mark 8. Other details of the experiment were exactly the same as in Procedure A.

Results and Discussion

The investigation was carried out by systematically varying the rate of flow of oxygen, time of combustion, range of temperature and the nature of filling, one at a time. It has been observed that the nature of the filling and temperature of the combustion played a significant role for better analytical data. The substances combusted on "simple band filling" at the conventional temperature $700 \pm 25°$ did not yield good results. The results improved significantly (see Table I) by raising the temperature to $900 \pm 25°$. Temperatures higher than $900 \pm 25°$ created practical difficulties due to clinkering of the filling. Contrastingly, the "combination filling" used in Procedure A did not yield better analytical values even by raising the temperature to $900 \pm 25°$. Table I shows the analytical data obtained from the "combination filling" (Column A)

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Table I. Combustion Results on Combination and Simple Band Fillings

Compound	Molecular Formula	Found, %		Calcd. %	Deviation, %	
		A*	B**		A*	B**
Friedelin[13]	$C_{30}H_{50}O$	C: 83.26	84.44	84.50	— 1.24	— 0.06
		H: 10.91	12.63	11.73	— 0.82	+ 0.90
Taraxerone[14]	$C_{30}H_{48}O$	C: 84.31	84.51	84.90	— 0.59	— 0.39
		H: 10.92	11.40	11.32	— 0.40	+ 0.08
Lupenone[15]	$C_{30}H_{48}O$	C: 83.52	84.77	84.90	— 1.38	— 0.13
		H: 10.71	11.56	11.32	— 0.61	+ 0.24
Taraxerol[16]	$C_{30}H_{50}O$	C: 83.61	84.38	84.50	— 0.89	— 0.12
		H: 11.96	11.84	11.73	+ 0.23	+ 0.11
β-Amyrin[17]	$C_{30}H_{50}O$	C: 82.33	84.48	84.50	— 2.17	— 0.02
		H: 11.43	12.19	11.73	+ 0.30	+ 0.46
α-Amyrin[16]	$C_{30}H_{50}O$	C: 83.24	84.13	84.50	— 1.26	— 0.37
		H: 11.75	12.37	11.73	+ 0.02	+ 0.64
Erythrodiol[18]	$C_{30}H_{50}O_2$	C: 78.44	79.00	79.00	— 0.56	± 0.00
		H: 11.60	11.63	11.11	+ 0.49	+ 0.52
Oleanolic acid[19] ...	$C_{30}H_{48}O_3$	C: 77.16	79.38	78.94	— 1.78	+ 0.44
		H: 10.54	10.80	10.50	+ 0.04	+ 0.30
α-Amyrin acetate[17]	$C_{32}H_{52}O_2$	C: 81.59	81.97	82.05	— 0.46	— 0.08
		H: 10.95	11.30	11.11	— 0.16	+ 0.19
β-Amyrin acetate[16]	$C_{32}H_{52}O_2$	C: 81.52	82.15	82.05	— 0.53	+ 0.10
		H: 11.57	11.92	11.11	+ 0.46	+ 0.81
Ursolic acid[20]	$C_{30}H_{48}O_3$	C: 78.41	78.84	78.94	— 0.53	— 0.10
		H: 10.89	10.80	10.50	+ 0.39	+ 0.30
Ursolic acid acetate[20]	$C_{32}H_{50}O_4$	C: 76.59	76.89	77.10	— 0.51	— 0.21
		H: 9.99	10.29	10.04	— 0.05	+ 0.25
Stigmastanol[21]	$C_{29}H_{52}O$	C: 82.50	83.42	83.65	— 1.15	— 0.23
		H: 12.77	12.88	12.50	+ 0.27	+ 0.38
Stigmastanone[21] ...	$C_{29}H_{51}O$	C: 83.00	83.78	83.85	— 0.85	— 0.07
		H: 12.40	12.35	12.29	+ 0.11	+ 0.06
Stigmasterol[21]	$C_{29}H_{48}O$	C: 83.60	84.18	84.46	— 0.86	— 0.28
		H: 11.81	11.84	11.65	+ 0.16	+ 0.19
β-Sitosterol[21]	$C_{29}H_{50}O$	C: 82.15	84.00	84.05	— 1.90	— 0.05
		H: 12.30	12.21	12.07	+ 0.23	+ 0.14
Wallichiene[22]	$C_{30}H_{50}$	C: 86.80	87.54	87.81	— 1.01	— 0.27
		H: 12.45	12.60	12.19	+ 0.26	+ 0.41
Isofernene[22]	$C_{30}H_{50}$	C: 86.44	87.03	87.81	— 1.37	— 0.78
		H: 12.40	12.25	12.19	+ 0.21	+ 0.06
Wallichiediene[22] ...	$C_{30}H_{50}$	C: 86.39	87.67	87.81	— 1.42	— 0.14
		H: 12.27	12.17	12.19	+ 0.08	— 0.02

* Procedure A.
** Procedure B.

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and "simple band filling" (Column B). The compounds analyzed were either of natural origin or derivatives of natural products.

It has been observed that the acetates yield better results with the improved conditions (Table I, Column B) which may be attributed to the thermal volatility of the compounds. Exception to this, however, are the triterpenoid ketones, e. g., lupenone, taraxerone and friedelin.

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Summary

An improved microcombustion technique for C-H-determination of steroids and triterpenoids using "simple band filling" at 900 ± 25° temperature is described.

Zusammenfassung

Eine verbesserte Mikroverbrennungstechnik zur Bestimmung von C und H in verschiedenen Steroiden und Triterpenen unter Verwendung einer einfachen Rohrfüllung bei 900 ± 25° wird beschrieben.

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