

Carboxymethoximes(I)

Carboxymethoximes and Their Methyl Esters of Ketosteroids

by

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카르복시메톡심(I)

케로스테로이드의 카르복시메톡심과 이들의 메틸 에스테르

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요 약

몇 가지의 ketosteroid의 carboxymethoxime 및 이들의 methyl ester를 합성하였고 carboxymethoxime의 중화 당량으로부터 ketosteroid의 분자량을 결정할 수 있음을 지적하였다. 또한 이들의 methyl ester는 ketosteroid의 화학인에 이용할 수 있음도 아울러 지적하였다.

ABSTRACT

The carboxymethoximes and their methyl esters of several ketosteroids were synthesized. It is suggested that the molecular weights of the ketosteroids could be determined through measuring the neutralization equivalents of their carboxymethoximes. The methyl esters of these carboxymethoximes could be utilized as useful derivatives for the identification of the ketosteroids.

Aminoxyacetic acid has been known having a bactericidal activity and employed as a preservative for proteins, vaccins and plasma.^(1,2) Its antibiotic activity is presumably due to its ability to combine with α -diketones, the most important in living bacteria being pyruvate.⁽³⁾

Aminoxyacetic acid reacts with carbonyl compounds refluxing in alcoholic solution and forms corresponding carboxymethoximes.⁽⁴⁾ Since the carboxymethoximes are soluble in aqueous alkali, it was suggested that

it could be used to separate ketosteroids from non-carbonyl compounds in oil.⁽⁵⁾ However, there have not been done much investigation upon these steroidal carboxymethoximes previously. This paper describes the synthesis of several steroidal carboxymethoximes and their properties. (Table 1)

The ketosteroids gave corresponding carboxymethoximes in good yield when they were refluxed in alcoholic solution of aminoxyacetic acid hemihydrochloride, $(\text{NH}_2\text{OCH}_2\text{COOH})_2 \cdot \text{HCl}$, and sodium acetate. When the rea-

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These were run without the presence of sodium acetate, the methyl esters of the corresponding carboxymethoximes, instead of the free acids, were obtained. Pregnenolone carboxymethoxime methyl ester was also obtained by the action of diazomethane on pregnenolone carboxymethoxime in methanol. The ester obtained from this reaction was identical in every aspect with the sample from the direct condensation. Progesterone, a dione, gave bis-carboxymethoxime on the normal condition.

The carboxymethoximes of α, β -unsaturated ketosteroids are much more soluble in alcohol than the parent ketones. However, pregnenolone carboxymethoxime is not very soluble in cold alcohol. It crystallizes out from the hot reaction mixture on cooling. Owing to this poor solubility in cold alcohol, the neutralization equivalent, when it was determined in aqueous methanol, was obtained as somewhat higher value. The methyl esters are less soluble in alcohol than the corresponding carboxymethoximes as expected. The carboxymethoximes,

except pregnenolone carboxymethoxime, melt in the range of 160-180° with decomposition. The darkening in color and gas evolution were observed with melting. Pregnenolone carboxymethoxime melts above 265° with decomposition. The gas evolution might be accounted to the decarboxylation at the carboxymethoxime side chain. The methyl esters melt at the temperature lower than the melting point of the parent ketosteroids without any apparent decomposition.

Since the acidity of the carboxymethoximes are strong enough to be titrated with dilute sodium hydroxide solution, the neutralization equivalents of these compounds could be determined very easily and accurately in the aqueous alcoholic solution. This provides a convenient and an accurate method to determine the molecular weights of the complex ketosteroids. The methyl esters, with their definite and considerably low melting points, could be used as the easily prepared derivatives for the identification of the ketosteroids.

Table 1. The carboxymethoximes and their methyl esters of ketosteroids

Ketosteroids	M.P.	Recrystallization solvent	Yield (%)	Analysis					
				Calcd.			Found		
				C	H	N.E.	C	H	N.E.
Pregnenolone	over 205 d.	Methanol	90	70.92	9.06	389.5	70.20	9.07	401.9
Methyl ester	185-6	Methanol	92	71.43	9.24	—	71.53	9.24	—
Progesterone (bis-)	190-2 d.	Ethyl acetate-ethanol	96	65.19	7.88	230.3	65.37	8.12	251.1
Dimethyl ester	83-5	Methanol	90	66.36	8.25	—	66.17	8.06	—
Testosterone	174-6 d.	Ethyl acetate-cyclohexane	96	69.77	8.65	361.5	69.57	8.66	357.7
Methyl testosterone	170-1 d.	aq. Methanol	95	70.37	8.86	375.5	70.38	9.18	377.5
Methyl ester	135-8	Benzene-ligroin	70	70.92	9.06	—	70.66	8.98	—
Ethisterone	164-5 d.	Ethyl acetate-cyclohexane	93	71.66	8.11	385.5	71.40	8.23	384.7
Methyl ester	141-2	Methanol	85	72.15	8.33	—	71.86	8.33	—

Experiments and Results

Almost identical procedures were utilized for the preparations of the ketosteroids and their methyl esters except pregnenolone, although somewhat varied amounts of the solvent were used for each compounds. For pregnenolone a slightly different procedure was used. Here, the typical procedures for the preparations of testosterone carboxymethoxime and its methyl ester, and pregnenolone carboxymethoxime and its methyl ester are given. The results from the experiments are summarized in Table 1.

Pregnenolone carboxymethoxime. Pregnenolone (2 g), aminoxyacetic acid hemihydrochloride (0.9 g),

and sodium acetate trihydrate (0.5 g) were refluxed in 50 ml of methanol for one half hour. After cooling the reaction mixture was diluted with water to 100 ml and filtered. The white precipitate was recrystallized from methanol. Yield, 2.2 g (90 %).

Testosterone carboxymethoxime. Testosterone (1 g), aminoxyacetic acid hemihydrochloride (0.6 g), and sodium acetate trihydrate (1.0 g) were refluxed in 15 ml of 90% ethanol for an hour. The reaction mixture was distributed between 100 ml of 1% potassium carbonate solution and 100 ml of ether. The aqueous layer was separated and washed with ether. The alkaline solution was neutralized with dilute hydrochloric acid using congo red as the indicator. The precipitate

was filtered and washed with water. Recrystallized from ethyl acetate-cyclohexane. Yield, 1.2 g (96 %).

Pregnenolone carboxymethoxime methyl ester.

Procedure A. Pregnenolone (2 g) and aminoxyacetic acid hemihydrochloride (0.8 g) were refluxed in 50 ml of methanol for one half hour. After cooling, deposited crystals were filtered and washed with methanol. Recrystallized from methanol. Yield, 2.3 g (92%). mp, 185-6°.

Procedure B. Pregnenolone carboxymethoxime (0.2 g) was dissolved in a small amount of methanol. A large excess of diazomethane solution in ether was added into this solution. The reaction mixture was allowed to stand at room temperature for overnight. Ether was removed from the colorless solution by evaporation. The crystals were filtered and washed with a small amount of methanol. Recrystallized from methanol. mp, 185-6°. Yield, 0.15 g.

The mixed sample of the above two preparations melted at the same temperature without depression. Identical infrared absorption spectra were also obtained.

Progesterone bis-carboxymethoxime dimethyl ester. Progesterone (1 g) and aminoxyacetic acid hemihydrochloride (0.8 g) were refluxed in 20 ml of methanol for one half hour. The reaction mixture was diluted with water (10 ml) and placed in an ice bath. The initially oily liquid which was separated out from the reaction mixture turned into crystalline lumps. This was filtered and washed with water. Recrystallized from methanol. Yield, 1.4 g (90 %).

References

- 1) C. B. Favour; *J. Bact.*, 55, 1 (1948)
- 2) *U. S. Patent*, 2,464,197
- 3) M. Anchel and R. Schoenheimer; *J. Biol. Chem.*, 114, 539 (1936)