4-1-1935

Some aldehyde condensation derivatives of creatinine

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SOME ALDEHYDE CONDENSATION DERIVATIVES OF CREATININE

THESIS

Presented in Partial Fulfillment of the Requirements for the Degree of Master of Science in the Graduate Department of the University of Richmond

by

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The University of Richmond

1935

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TABLE OF CONTENTS

	Page
1. Introduction	3
2. History	5
3. Experimental	7
4. Discussion of Results	10
5. Summary	14
6. Acknowledgement	15
7. Autobiography	16

INTRODUCTION

When insulin, a substance which is used to reduce sugar in the blood, is hydrolysed, products are obtained bearing some similarity to creatinine.

Jensen and Wintersteiner, J. Biol. Chem., 98, 281-7 (1932)

Therefore, reasoning conversely, creatinine derivatives might be expected to possess a hypoglycemic (sugar reducing) effect.

Jordan

M. S. Thesis, University of Richmond, 1934

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prepared some derivatives of creatinine which were fed orally to rabbits at the Medical College of Virginia and were reported to have no effect on blood sugar. Lazarus and Snellings

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M. S. Theses, University of Richmond, 1935

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continued this work but the derivatives which they prepared have not been tested at this time.

Jordan discovered while working on aldehyde condensations of creatinine that a by-product could be isolated which was

lower in nitrogen content than could be explained on the basis of one molecule of the aldehyde condensing with one of creatinine. On the basis of the nitrogen analysis and the fact that the furfural derivative gave furfural on treatment with concentrated hydrochloric, he described these compounds as derivatives formed by the condensation of two aldehyde molecules with one of creatinine.

The object of the present investigation is to study these aldehyde condensations with special regard to the best methods of preparation and the isolation and purification of any by-products. It is intended at some future date to subject all the derivatives to tests for hypoglycemic activity.

HISTORY

Creatine was reported in 1832 by Chevreul. It is probable that he also discovered creatinine at this time. After a great deal of work on its structure, an excellent outline of which one will find in Hunter's monograph on creatine and creatinine,

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Creatine and Creatinine by Andrew Hunter- Longmans, Green & Co.

Strecker in 1867 proposed that creatine was methyl guanidine acetic acid and that creatinine was its internal anhydride. This view has continued to the present day.

Using the method of Erlenmyer Jr.,

Ann., 284, 49 (1895)

Nicolet and Campbell

prepared benzalcreatinine.

J. A. C. S., 50, 1155 (1928)

Richardson, Welch, and Calvert

J. A. C. S., 51, 3075 (1929)

by fusing aldehydes with creatinine prepared 5-m-nitrobenzalcreatinine and 5-(m-methoxy-p-hydroxybenzal)-creatinine. Cornthwaite and Jordan

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J. A. C. S., 56, 2733 (1934)

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prepared by the method of Richardson, 5-furfuralcreatinine, difurfuralcreatine, 5-furfural-methylcreatinine, di-(furfural-acrolein)-creatinine, 5-salicylcreatinine, 5- cinnamylcreatinine, dicinnamylcreatinine, and difurfuralmethylcreatinine

EXPERIMENTAL

5-Piperonalcreatinine: 3 grams of creatinine and 8 grams of piperonal were heated at 180-90 degrees C. on an oil bath for one hour or until it is finished reacting. After ether extraction, the mass was boiled up with successive portions of hot water. On cooling, fine yellow needles separated. This compound is insoluble in cold water, acetone, and ether, but soluble in hot water and hot alcohol. M.P. 274 (closed tube). The yield was 2 grams (31%). Analysis. Calculated for $C_{12}H_{11}O_3N_3$: N, 17.14. Found: N, 16.55

note- Since all of these compounds melt rather indistinctly and with decomposition, their recorded melting point may vary with the observer. The point generally taken was the point at which the solid collapsed forming a dark liquid, sometimes with effervescence. It was thought that a sealed tube gave a sharper melting point and so all melting points were taken in that manner.

5-Piperonalcreatinine picrate: This compound was formed when an aqueous solution of picric acid was added to an acetic acid solution of piperonalcreatinine. Recrystallization from hot water gave a M.P., 255.

Tripiperonaldicreatinine: After extracting the reaction product of the piperonal condensation with hot water, there was left a reddish orange residue, which was then extracted with glacial acetic acid in which it was only slightly soluble, and then recrystallized from hot aniline. This compound was characterized by its great insolubility in the common solvents, being insoluble in dilute hydrochloric acid, dilute potassium hydroxide, benzene, ether, alcohol, and only slightly soluble in glacial acetic acid.

M.P., 327; .3 grams (4%).

Analysis. Calculated for $C_{32}H_{26}O_8N_6$: N, 13.50. Found: N, 13.88

5-o-chlorobenzalcreatinine: 5 grams of creatinine and 7 cc of o-chlorobenzaldehyde were heated at 140-50 for one hour. After extraction with ether, the fused mass was boiled with dilute hydrochloric acid, filtered and the filtrate neutralized with ammonia. The precipitate was filtered and washed with hot water. This compound consisted of yellow needles which became almost white on continued purification. It is slightly soluble in alcohol, acetone, and hot water. M.P., 242; yield, 6 grams (57%).

Analysis. Calculated for $C_{11}H_{10}ON_3Cl$: N, 17.87. Found: N, 17.31.

5-o-chlorobenzalcreatinine picrate: From alcohol it melted at 260.

5-o-chlorobenzalcreatinine hydrochloride: 1 gram of o-chlorobenzalcreatinine was dissolved in concentrated hydrochloric acid. On cooling, shiny yellow needles separated. These were crystallized from concentrated hydrochloric acid. The compound is soluble in alcohol and water. M.P., 241; yield .7 grams (61%).

Analysis. Calculated for $C_{11}H_{11}ON_3Cl_2$: N, 15.49. Found: N, 15.34.

Tri-o-chlorobenzaldicreatinine: 5 grams of creatinine and 10.6 cc of o-chlorobenzaldehyde were heated to 180-90. The mass was then extracted with ether and then with hot water to remove the monocompound. The residue was then crystallized from acetic acid. The compound consisted of yellow crystals. Subsequent purification gave a M.P., 270.

Analysis. Calculated for $C_{29}H_{23}O_2N_6Cl_3$: N, 14.17. Found: N, 14.26.

Tri-o-chlorobenzalcreatinine picrate: From acetic acid it melted at 270. A mixed melting point proved that it was not tri-

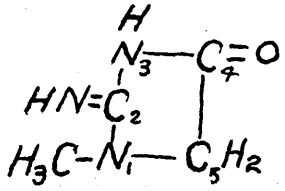
o-chlorobenzaldicreatinine.

Creatinine Pyruvate: 5 grams of creatinine and 6.3 cc of pyruvic acid were heated for 45 minutes at 112-20. The product was then extracted with ether and crystallized from alcohol. The product was then washed with ether, dryed in a vacuum desiccator, and then in an oven at 110. This compound consisted of white microscopic crystals. Titration with alkali gave a molecular weight of 213 (calculated, 201). The compound was very soluble in water. M.P. 178 (effervescent without charring).

Analysis. Calculated for $C_7H_{11}O_4N_3$: N, 20.89. Found: N, 20.92.

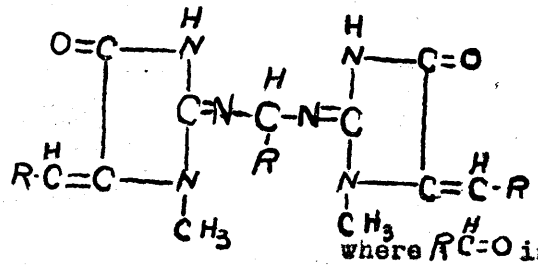
DISCUSSION OF RESULTS

The following notation which is used in Chemical Abstracts will be used in this discussion.



Condensation Temperatures: The most favorable temperatures for the condensation of these aldehydes was between 140-50. Higher temperatures appeared to favor the production of polyaldehyde derivatives.

Side Products: Every time the piperonal condensation was carried out, there was a small residue left of orange-red color, whose nitrogen content corresponded to a condensation product of three aldenyde molecules with two of creatinine. A possible formula for this type of compound is



where $R\overset{H}{C}=O$ is the aldehyde used. While this formula is merely hypothicated, the fact that it is insoluble in almost all of the solvents tried would lend support to its very complex nature.

The o-chlorobenzaldehyde condensation also produced two compounds. On removing the mono-derivative with hot water or dilute

hydrochloric acid there was left a residue which was crystallized from glacial acetic acid and gave a nitrogen analysis which led one to believe that it was a tri-di derivative similar to the piperonal derivative formerly described. The picrate of this compound was prepared and characterized.

The Action of Concentrated Hydrochloric Acid on Tri-o-chlorobenzaldicreatinine: The compound was refluxed with concentrated hydrochloric acid for 8 hours. On filtering, 80% of the original compound was recovered. The hydrochloric acid on cooling gave no appreciable precipitatebut on making alkaline a flocky precipitate came down whose melting point was above 315 and could not be identified. When the filtrate was extracted with ether, no appreciable residue was found. This experiment was performed because Jordan had been able to hydrolyze off the second aldehyde group of difurfuralcreatinine. The fact that this compound was so extremely stable toward acid confirms the belief that this may be a new type of creatinine derivative. In trying to repeat Jordan's hydrolysis of difurfuralcreatinine the compound went into solution readily enough but the mono-derivative could not be isolated from the reaction mixture. However, the experiment was not repeated and no doubt a refinement of technique would have led to the identification of the reaction products. When dicinnamylcreatinine was treated with concentrated hydrochloric acid, a small amount of the picrate of 5-cinnamylcratinine was isolated as was a small amount of a substance which had the characteristic smell of cinnamic aldehyde. However, the amounts of both substances which were isolated were

so small that it is quite possible that they were both present as impurities in the original dicinnamylcreatinine. There was left a considerable residue which could not be dissolved in the acid.

Attempted Condensation with Pyruvic Acid: Since pyruvic acid has a carbonyl group and has been shown to be active in condensations, an attempt was made to condense this compound with creatinine. The reaction product, when crystallized from alcohol gave a nitrogen analysis which corresponded to an addition product of the two molecules. From the evidence it seemed quite probable that this compound was creatinine pyruvate.

$$C_4H_7ON_3 \cdot C_3H_4O_3$$

The compound was titrated with alkali and its molecular weight found to be 213(calculated, 201). When this solution was evaporated, extracted with absolute alcohol, creatinine was obtained as was its picrate on the addition of picric acid. Creatinine pyruvate is very soluble in water.

Attempted Condensation of Pyruvic Acid with Piperonal Creatinine: Since pyruvic acid has been shown to condense thus:

$$R\overset{H}{C}=O + H_2\overset{H}{C}-\overset{O}{\overset{\|}{C}}-COOH \xrightarrow{-HOH} R\overset{H}{C}=\overset{H}{C}-\overset{O}{\overset{\|}{C}}\cdot COOH$$

it was thought that this reaction might take place on the carbonyl group of creatinine or that the carbonyl group of pyruvic acid might condense on the imino group of creatinine as it is thought that furfural does. Piperonalcreatinine and pyruvic acid were heated, but, no derivative of creatinine could be obtained, approximately one-half of the original compound being recovered. Its melting point and that of its picrate checked. It is probable that the ethyl ester of pyruvic acid or sodium pyruvate would be more likely to condense.

Action of Sodium on Creatinine: It was thought that the methylene group in creatinine might be reactive toward sodium analagously to the malonic ester synthesis. Accordingly, creatinine was suspended in absolute alcohol, sodium added, and then ethyl iodide. After refluxing, the mixture was filtered, and when the solution cooled, white crystals appeared. An analysis proved it to be creatinine.

SUMMARY

1. The following compounds have been prepared and described:

5-Piperonalcreatinine and its picrate

Tripiperonaldicreatinine

5-o-chlorobenzalcreatinine and its picrate

5-o-chlorobenzalcreatinine hydrochloride

Tri-o-chlorobenzaldicreatinine and its picrate

Creatinine pyruvate

2. Tri-o-chlorobenzaldicreatinine was found to be stable towards concentrated hydrochloric acid.

3. Structures have been proposed for tripiperonaldicreatinine and tri-o-chlorobenzaldicreatinine.

4. Creatinine did not react with ethyl iodide in the presence of sodium and absolute alcohol.

5. Pyruvic acid did not condense with creatinine or piperonal creatinine.

ACKNOWLEDGEMENT

The author wishes to take this occasion to acknowledge his debt to Dr. W. R. Cornthwaite, under whose direction this research was carried on.

He also wishes to thank the Valentine Meat Juice Co. who furnished the creatine necessary in this investigation.

AUTOBIOGRAPHY

Clarence England Denoon Jr. was born February 25, 1915, in Richmond, Virginia; attended the public schools of Richmond, and graduated from John Marshall High School in February, 1931. He attended the University of Richmond three years and two summers, received the degree of Bachelor of Science in June, 1934 and in September, 1934 matriculated as a graduate student in the Graduate Department of the University of Richmond.