

**Absorption.** Dabei wird die Oberfläche allmählich grau bis schwarz oder farbig, die Helligkeit ist aber vom Beobachtungswinkel unabhängig. 2. *Änderung durch zunehmende Spiegelung.* Fällt das Licht z. B. unter  $45^\circ$  ein, so nimmt mit zunehmendem Glanz die Helligkeit bei senkrechter Aufsicht ab, unter dem Spiegelwinkel aber zu, bis schließlich im extremen Fall die Helligkeit bei Aufsicht auf Null sinkt, unter dem Spiegelwinkel aber die des einfallenden Lichtes erreicht. 3. *Änderung*

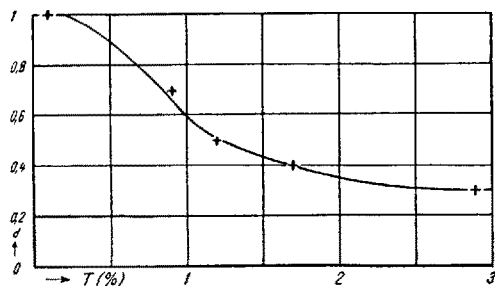


Abb. 3. Durchlässigkeit der Barytplatten  
 $T$  = Durchlässigkeit in % bei  $555\text{ m}\mu$   
 $d$  = Schichtdicke in mm

*im Falle zunehmender Durchsichtigkeit.* In dem Maße, wie die Oberfläche durchscheinender wird und das Licht mehr in die Tiefe dringt, nimmt die Helligkeit bei Aufsicht unter jedem Winkel immer mehr ab, in der Durchsicht aber immer mehr zu, bis schließlich im Idealfall alles auffallende Licht, ähnlich wie bei einem nichtspiegelnden Glas, die Oberfläche durchdringen würde. Praktisch findet meist eine Überdeckung der drei Fälle statt. Der Grad der Durchsicht einer weißen Oberfläche ergibt sich übrigens aus der Deckkraft, die angibt, bei welcher Schichtdicke senkrecht auffallendes Licht auf z. B. 1 % geschwächt wird. In unserem Falle ergab die Messung 0,6 mm für Licht von  $555\text{ m}\mu$  Wellenlänge, vgl. Abb. 3. Selbstverständlich hängt die Deckkraft noch von der Wellenlänge ab; sie ist im blauen Spektralbereich etwa 2,5mal größer als im roten.

K. MIESCHER und R. ROMETSCH

Forschungslaboratorien der Ciba Aktiengesellschaft, Basel, den 28. April 1950.

### Summary

Precipitation of bariumsulfate with carefully purified reactives under carefully controlled conditions to avoid any contamination, and pressing of this barite in chromium-plated moulds with a trace of gelatine yields *white standards* of almost ideal properties: the high reflectance of .991 varies only  $\pm 0,002$  throughout the visible spectrum and practically no gloss can be observed.

## The Deamination of Djenkolic Acid in Rats

A naturally occurring amino acid of the djenkol bean, named djenkolic acid (VAN VEED and HYMAN<sup>1</sup>), was synthesized by the direct combination of formaldehyde with L-cysteine by ARMSTRONG and DU VIGNEAUD<sup>2</sup>. The

structure of this compound, which is principally formed by two condensed molecules of cysteine, gave us the impulse for the investigation of its action in the animal body. The deamination *in vivo* was evaluated by the determination of the urinary total nitrogen and amino acid nitrogen, and of the level of non-protein nitrogen in blood. The second question would be whether the djenkolic acid has a glycogenic action.

**Experimental.** We have synthesized the djenkolic acid according to the directions of ARMSTRONG and DU VIGNEAUD (l. c.) from L-cysteine hydrochloride Roche. The monohydrochloride of this product has been prepared and used for experiments. It exhibited a melting point of  $265^\circ\text{C}$  (with decomposition);  $[\alpha]_D^{20.5} = -61 \pm 1^\circ$  for a 1 per cent solution in 1 N HCl. The compound is slowly soluble in water, and the dose given to the animals by mouth was not completely absorbed from the intestine, as was shown by trials.

For studying the urinary excretion of total nitrogen and amino acid nitrogen, each rat was put in an individual metabolism cage, fasted, and the urine was twice collected in the course of 48 hours; after the first 24 hours the substance under examination was given by a gastric tube. Total nitrogen was determined by micro-Kjeldahl method (steam distillation) and estimated titrimetrically, free amino acid nitrogen by the formol titration method (HENRIQUES and SØRENSEN<sup>1</sup>). The glycogenic action was evaluated according to the glycogen content of the liver. Young male albino rats weighing 105–125 g were always fasted for 24 hours before the experiment. Thereafter the substance to be examined was given with the aid of a gastric tube. The period allowed for glycogen formation was 3 hours, the same as that recommended by SCHOFIELD and LEWIS<sup>2</sup>, to avoid any complicating factors. After this period the animals were killed and the liver glycogen isolated by a micro-modification of the PFLÜGER's<sup>3</sup> method. After hydrolysis of the glycogen, the glucose obtained was estimated by the ferrocyanide procedure of FOLIN<sup>4</sup> with the aid of the Coleman photometer.

The non-protein nitrogen in blood was determined 3 hours after the administration of the examined substance by the micro-Kjeldahl method with steam distillation, estimations were made by direct nesslerization.

In all the groups beside djenkolic acid, experiments with L-cysteine and D-alanine on the one hand and control analyses of normal animals on the other hand were run in parallel.

The results of individual experiments are summarized in Tables 1 and 2. The nitrogen excretion indicates that djenkolic acid should be deaminized in rats. The extent of this reaction is however less than with cysteine, in consequence of the poor solubility of this acid. Also the increase of non-protein nitrogen in the blood after administration of djenkolic acid, which is smaller than after treatment with cysteine, corresponds to it. The use of longer periods of absorption would most probably be desirable for quantitative comparison. The increased amount of amino acid nitrogen in urine was similar to that occurring after the administration of D-alanine, but smaller than after cysteine. Also the measurement of the glycogen formation in fasting rats shows that the utilization of djenkolic acid in respect to gluconeogenesis seems to be of a magnitude which is not quite equal to the utilization of cysteine.

Therefore it cannot be suggested that in rats djenkolic acid is split into its two cysteine-halves and metabolized via this last compound. DYER<sup>5</sup> reported coincidentally that feeding of djenkolic acid could not support growth

<sup>1</sup> V. HENRIQUES and S. P. L. SØRENSEN, *Z. physiol. Chem.* **64**, 120 (1910).

<sup>2</sup> F. A. SCHOFIELD and H. B. LEWIS, *J. Biol. Chem.* **169**, 373 (1947).

<sup>3</sup> E. PFLÜGER, *Arch. Physiol.* **96**, 1 (1903).

<sup>4</sup> O. FOLIN, *J. Biol. Chem.* **77**, 421 (1928).

<sup>5</sup> H. M. DYER, *J. Biol. Chem.* **119**, xxviii (1937).

<sup>1</sup> A. G. VAN VEED and A. J. HYMAN, *Rec. Trav. chim. Pays-Bas* **54**, 493 (1935).

<sup>2</sup> M. D. ARMSTRONG and V. DU VIGNEAUD, *J. Biol. Chem.* **168**, 373 (1947).

Table I  
Urinary excretion of Total Nitrogen and Amino Acid Nitrogen

| No. of rats | Agent               | Excreted             | Per 24 hours<br>before administration |             |             | Per 24 hours<br>after administration |             |             | Difference          |
|-------------|---------------------|----------------------|---------------------------------------|-------------|-------------|--------------------------------------|-------------|-------------|---------------------|
|             |                     |                      | Mean ( $\bar{X}$ )                    | S†          | SM††        | Mean ( $\bar{X}$ )                   | S           | SM          |                     |
| 20          | None                | T-N*, mg<br>A-N**, % | 87.8<br>2.9                           | 13.2<br>0.8 | 4.2<br>0.3  | 80.0<br>3.5                          | 18.3<br>1.1 | 5.8<br>0.3  | - 8.9%<br>+ 21.0%   |
| 12          | Djenkolic Acid 2 mM | T-N, mg<br>A-N, %    | 94.3<br>3.3                           | 26.4<br>1.4 | 10.8<br>0.6 | 110.5<br>9.5                         | 22.7<br>1.9 | 9.3<br>0.8  | + 17.2%<br>+ 188.0% |
| 6           | L-Cysteine 2 mM     | T-N, mg<br>A-N, %    | 97.8<br>3.0                           | 26.4<br>0.9 | 15.3<br>0.5 | 144.0<br>23.0                        | 21.4<br>2.1 | 12.4<br>1.2 | + 47.2%<br>+ 667.0% |
| 6           | D-Alanine 2 mM      | T-N, mg<br>A-N, %    | 84.2<br>3.4                           | 9.5<br>0.2  | 5.5<br>0.1  | 111.5<br>10.1                        | 4.2<br>1.2  | 2.4<br>0.7  | + 32.4%<br>+ 197.0% |

\* mg of total nitrogen in urine.

\*\* Per cent of total nitrogen excreted as amino acid nitrogen.

† Standard deviation,  $S^2 = \frac{\sum (X-\bar{X})^2}{N-1}$ †† Standard error,  $S_M^2 = \frac{S^2}{N}$ 

Table II  
Glycogen Content of Liver and Non-protein Nitrogen Content of Blood after 3-Hour Period

| No. of rats | Agent               | Analysed              | Maximum       | Minimum       | Mean ( $\bar{X}$ ) | S            | SM           | P†                      |
|-------------|---------------------|-----------------------|---------------|---------------|--------------------|--------------|--------------|-------------------------|
| 20          | None                | *Gl., %<br>**N-N, mg% | 0.202<br>39.4 | 0.059<br>26.9 | 0.113<br>33.0      | 0.042<br>3.5 | 0.009<br>0.7 |                         |
| 16          | Djenkolic Acid 1 mM | Gl., %<br>N-N, mg%    | 0.386<br>43.6 | 0.124<br>28.3 | 0.221<br>35.1      | 0.089<br>3.9 | 0.022<br>0.9 | $\leq 0.01$<br>$= 0.05$ |
| 6           | L-Cysteine 1 mM     | Gl., %<br>N-N, mg%    | 0.357<br>—    | 0.127<br>—    | 0.190<br>—         | 0.095<br>—   | 0.039<br>—   | $= 0.05$<br>—           |
| 7           | D-Alanine 1 mM      | Gl., %<br>N-N, mg%    | 0.522<br>40.3 | 0.154<br>32.1 | 0.318<br>37.2      | 0.137<br>3.2 | 0.052<br>1.2 | $< 0.01$<br>$< 0.01$    |

\* Glycogen content of liver, per cent.

\*\* Non-protein nitrogen of blood, mg per 100 ml.

† Probability of error, according to FISCHER's "Statistical Methods for Research Workers".

on a cysteine deficient diet. The interaction of many factors is most probably concerned, and meanwhile it is difficult to form from the present exploratory experiments an idea of the individual intermediates in djenkolic acid metabolism.

JAROSLAV RERÁBEK

Department of Biology, Faculty of Medicine, Charles' University, Prague, March 6, 1950.

#### Zusammenfassung

Es wurde die Desaminierbarkeit und die Glukoplastizität der nach ARMSTRONG und DU VIGNEAUD synthetisch hergestellten Djenkolsäure bei Ratten geprüft. Der Gesamt-N-Ausscheidung nach kann der Stoff im Rattenkörper desaminiert werden und bewirkt dabei eine Erhöhung des Leberglykogens. Beide Reaktionen sowie die Ausscheidung des Aminostickstoffs und die Menge des Nichtweiß-N im Blute, haben jedoch ein geringeres Ausmaß als nach Einverleibung von Zystein. Danach scheint es, daß das Molekül der Djenkolsäure

nicht einfach in seine beiden Zysteinhälften zerfällt und auf diesem Wege umgewandelt wird, sondern es muß mit einem komplizierteren Umbau gerechnet werden.

#### Métabolisme de l'adrénochrome et du trihydroxyméthylindole chez le lapin

Le sort que subit l'adrénaline au cours de son métabolisme normal dans l'organisme a fait l'objet de nombreuses recherches et toutes les possibilités d'attaque de sa molécule ont été envisagées<sup>1,2</sup>.

L'étude de l'oxydation *in vitro* de l'adrénaline a fait de nombreux progrès et on peut, semble-t-il aujourd'hui, établir les différents stades de transformation de l'adrénaline jusqu'au stade terminal d'oxydation générale-

<sup>1</sup> Z. M. BACQ, J. Pharm. Exp. Therap. 55, (II), 1 (1949).

<sup>2</sup> Z. M. BACQ, P. FISCHER et J. LECOMTE, Bull. Soc. Chim. biol. 31, 395 (1949).