

showed less distortion and apparently less maceration had occurred than in comparable cases of retention *in utero*. These fetuses felt quite firm in contrast to the soft yielding fetal mass generally found when fetuses of this size were retained *in utero*. The placentæ were firm and appeared quite shrunken.

In another experiment of the same nature 6 fetuses measuring 32 mm. were placed in the body cavity. The embryonic membranes were torn and folded off these fetuses but in each case were placed in the body cavity with the placentæ. This female seemed healthy and normal; eating well and appearing quite lively. The retained dead fetuses had no apparent effect upon the mother. She was sacrificed and autopsied after 21 days. The appearance of the fetuses and their placentæ was similar to that of those just described.

It appears from the above that retention of near term dead fetuses *in utero* or in the peritoneal cavity does not necessarily result in death of the mother.

### 8621 P

#### Gasometric Determination of Carboxyl Groups in Amino Acids.

DONALD D. VAN SLYKE AND ROBERT T. DILLON.

*From the Hospital of the Rockefeller Institute for Medical Research, New York.*

Ruhemann<sup>1</sup> and Grassmann and von Arnim<sup>2</sup> have shown that the color-forming reaction of ninhydrin with amino acids occurs with evolution of CO<sub>2</sub>. We have found that at pH 5, or somewhat lower, in water solution the CO<sub>2</sub> from carboxyl groups adjacent to alpha-amino groups can be quantitatively split off in 3 minutes. The apparatus used is that employed by Van Slyke, Page and Kirk<sup>3</sup> for manometric determination of organic carbon. In the combustion tube of this apparatus are placed a few milligrams of amino acid, 100 mg. of KH<sub>2</sub>PO<sub>4</sub>, and 50 mg. of ninhydrin made to a volume of 1.0 cc. with water. The tube is connected to the manometric chamber, arranged as for combustions,<sup>3</sup> and the solution is boiled gently for 3 minutes. The evolved CO<sub>2</sub> is measured as in carbon combustions.<sup>3</sup> The reaction appears to be specific for car-

<sup>1</sup> Ruhemann, S., *J. Chem. Soc.*, 1911, **99**, 792, 1486.

<sup>2</sup> Grassmann, W., and von Arnim, K., *Annalen der Chem.*, 1934, **510**, 288.

<sup>3</sup> Van Slyke, D. D., Page, I. H., and Kirk, E., *J. Biol. Chem.*, 1933, **102**, 635.

boxyl groups of amino acids. Other organic acids, such as acetic, lactic, citric, yield no  $\text{CO}_2$ . Each of the naturally occurring amino acids including proline yields 1 mole of  $\text{CO}_2$ , except the dicarboxylic acids, aspartic and glutamic. Aspartic yields at once 2 moles of  $\text{CO}_2$ , while glutamic yields one mole at once, and a second much more slowly (only about 2% in 3 minutes). Peptides do not react, even with the free carboxyl, which is inactivated by the binding of the alpha  $\text{NH}_2$  group in peptide linking. Urea does not react.

Because other primary amines, urea and ammonia do not interfere, the determination of amino acid carboxyl  $\text{CO}_2$  may be more specific for amino acids in biological material than is the nitrous acid reaction. Because of the non-reactivity of the free carboxyl group in peptides the method appears to provide a sharp distinction between peptides and free amino acids.

### 8622 C

#### Extraction of Gonadotropic Factors from the Blood. Improved Technic.

ROBERT T. FRANK AND U. J. SALMON.\*

*From the Laboratories of The Mount Sinai Hospital, New York City.*

An improved method for gonadotropic hormone (G.H.) extraction from blood was reported from this laboratory in 1935.<sup>1</sup> By means of this method, both follicle stimulating as well as luteinizing effects were obtained with 40 cc. of blood taken from normal menstruating women, from those past the physiological menopause as well as from surgical castrates.<sup>2</sup> This method, which involved desiccation of the blood with sodium sulphate, was used because it enabled both G.H. and E.S. (estrogenic substance) determinations to be performed on the same specimen.

By the present method the deleterious effect of alcohol upon the G.H. factors is avoided, giving a more complete G.H. extraction without impairing the E.S. yield. Moreover, the extracts obtained

---

\* Hiram N. Vineberg Research Fellow.

<sup>1</sup> Salmon, U. J., and Frank, R. T., *PROC. SOC. EXP. BIOL. AND MED.*, 1935, **32**, 1236.

<sup>2</sup> Frank, R. T., Salmon, U. J., and Friedman, R., *PROC. SOC. EXP. BIOL. AND MED.*, 1935, **32**, 1666.