yellowish colour of the split produkt can be measured photometrically at an optimal wavelength of 405 nm. Inactivation of thrombin by antithrombin III results in inhibition of this reaction. There is a linear relation between the percentage of thrombin inhibited and the activity of antithrombin III. As a result of test series on various modifications of the method an optimated procedure is described. The standard error of the assay was calculated at 3,2 per cent. Comparable results could be obtained between the coagulation method of Hensen and Loeliger and the chromogenic substrate method when both assays were carried out simultaneously on 18 patients as well as on pooled standard plasma.

Janet L. Lane, Prudence Bird and C. R. Rizza (Research Laboratory, Oxford Haemophilia Centre, Churchill Hospital, Oxford, England): **A New Assay for the Measurement of Total Progressive Antithrombin.** **(94)**

A new rapid method for assaying total antithrombin activity has been developed based on the inactivation of thrombin incorporated into an agarose gel, during the radial diffusion of plasma in the gel. The area of thrombin inactivation is subsequently observed by the coagulation of fibrinogen in a separate agarose gel layer poured over the thrombin gel. The method is described in detail and its accuracy assessed with respect to other antithrombin assays. Using specific antisera to α_2-globulin (antithrombin III), α_2-macroglobulin and α_1-antitrypsin, total antithrombin activity measured by this assay consisted of 47% α_2-globulin, 29% α_2-macroglobulin and 26% α_1-antitrypsin.

H. Nagasawa, Y. Hotta, N. Takara, M. Fujimaki and K. Fukutake (Department of Clinical Pathology, Tokyo Medical College, 7–1, 6-Chome, Nishi-Shinjuku, Shinjuku-Ku, Tokyo, Japan): **Inhibitory Effects of Estrogens on Antithrombin III.** **(95)**

After the addition of purified estrogen preparation to the plasma, the anticoagulant activity of antithrombin III measured by Bigg's method decreases in proportion to the amount of estrogen added. And the biological activity of antithrombin III in the plasma mixed with estrogen decreases proportionally in single immunodiffusion assay with anti-antithrombin III serum. The same findings mentioned above are also observed in the experiences with estron, equilin, equilenin and estradiol.

Furthermore, the rocket formation of antithrombin III fraction prepared by Abildgaard's method in crossimmunoelectrophresis also disappeares by the addition of estrogen into the reaction mixture, and the electric mobility of antithrombin III fraction with estrogen increases the migration to the anode in polyacrylamide gel electrophoresis.

U. Abildgaard, M. Lie, A. Teien and O. R. Ødegård (Medical Department A, Aker Hospital, Oslo 5, Norway): **Coagulation Inhibitors Studied with Chromogenic Substrate.** **(96)**

The rapid amidolysis of the chromogenic substrate Bz-Phe-Val-Arg-pNA by thrombin (Svendsen & al. Thromb. Res. *1*, 267, 1972) provides a useful tool for the study of coagulation inhibitors. With *purified reagents*, reversible inhibition of the reaction can be demonstrated with 0.01 U/ml of heparin. The inactivation of thrombin by antithrombin III (At-III) is accelerated by 0.001 U/ml of heparin, making it possible to assay small amounts of heparin. The amidolytic assay system has also been used to study the competition between thrombin and factor Xa for At-III.

The sensitivity and specificity of rapid amidolytic methods for assay of the following *plasma activites* will be reported:

1) At-III activity (contaminating heparin neutralized).
2) Exogenous heparin (during heparin treatment).
3) Combined effect of At-III and exogenous heparin.
4) At-III in the presence of optimal concentrations of heparin.