A rapid and simple method for estimating fibrinogen

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It is important during intravascular fibrinolytic therapy to check on the leakage of activator into the general circulation and the consequent breakdown of clotting factors. Serial fibrinogen estimations provide an important measure of any overdosage and serve as a guide to the amount of fibrinogen that may be required for replacement therapy and the response to such an infusion.

PRINCIPLE

In estimating the fibrinogen concentration it is preferable to measure what coagulates rather than what precipitates at a defined salt concentration. The principle of the test, therefore, is to convert the fibrinogen in citrated plasma into fibrin by the addition of calcium ions. The opacity of the clot so formed is then measured in an absorptiometer (colorimeter) using violet light. Assuming that all the fibrinogen is converted into fibrin the opacity of the clot is related to the fibrinogen concentration in the plasma (Rothnie, Norman, Steele, and Kinmonth, 1960).

METHOD

Blood samples were citrated and centrifuged to produce platelet-poor plasma. Equal volumes of this plasma and M/40 calcium chloride were mixed and placed in small cuvettes. These, though less accurate than larger cuvettes, were more economical of plasma. An initial reading was taken in the absorptiometer and represented the absorption of violet light due to the plasma mixture. The cuvettes were then incubated at 37°C. for about 15 minutes to allow a uniform fibrin clot to form. Readings were taken over the next five minutes until a constant value was obtained. The difference between the

constant value and the initial reading represented the opacity due to the fibrin clot. The addition of small amounts of thrombin to the plasma-calcium chloride mixture to accelerate the clotting was unsatisfactory because of the fine flocculation that readily occurred.

Any delay in the execution of the estimations necessitated keeping the blood or plasma samples on ice to limit fibrinogenolysis. The observation of the incubated clot measured in a crude way the rate of fibrinolysis.

CALIBRATION OF ABSORPTIOMETER

Pooled platelet-poor citrated plasma from women in the last trimester of pregnancy contains a high fibrinogen concentration and was used to calibrate the absorptiometer. Some of the plasma was carefully defibrinated by adding small quantities of thrombin and this was used to provide serially diluted samples of the pooled plasma. The opacities of the clots in these samples were measured and the fibrinogen concentration in each of them was estimated by the clot weight method (Ingram, 1952). A calibration graph was then constructed.

CONCLUSION

This method of estimating the fibrinogen concentration in plasma has the advantage of being simple and rapid and accurate enough for practical purposes.

REFERENCES

- Ingram, G. I. C. (1952). *Biochem. J.*, 51, 583.
Rothnie, N. G., Norman, A. G., Steele, M., and Kinmonth, J. B. (1960). *Brit. J. Surg.*, 48, 272.

DR. FEARNLEY asked if the opacity was affected by lipaemia.

MR. ROTHNIE replied that it is, but pointed out that all his patients had been starved for operation and had crystal clear plasma.