fixing time. By using the time-orientated relationships between adhesion and agitation and between aggregation and fixing time, it is possible to investigate platelet adhesion to collagen, platelet aggregation to collagen adhering platelets, and the relationship of the two.

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# A comparison of serum folate estimations using two different methods

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Most service laboratories have followed the method of Waters and Mollin (1961) for the routine estimation of serum folate. Herbert (1966) developed an 'aseptic' technique which obviated protein precipitation and sterilization necessary in the routine method but involved instead the collection of sterile serum. Recently a chloramphenicol-resistant strain of *Lactobacillus casei* has been developed for use in a fully automated system (Davis, Nicol, and Kelly, 1970). We report a comparison of serum folate results using a routine and a simple semi-automated 'chloramphenicol' method.

## Methods

### 'CHLORAMPHENICOL' SERUM FOLATE ASSAY

Stock cultures of a chloramphenicol-resistant strain of *L. casei* (NCIB 10463) were stored at 4°C in sterile lactobacillus broth AOAC (Difco) containing 100 μg/ml chloramphenicol BP. Twenty-four hours before assay the stock culture was subcultured into lactobacillus broth containing 10 μg/ml chloramphenicol. A dilute suspension of the washed organism was added to the cooled bulk medium.

On the day of assay, using micropipettes, 30 and 50 μl serum were each added to two tubes containing 4·0 ml half-strength inoculated BBL assay medium which contained 50 μg/100 ml ascorbic acid and 1·0 mg/100 ml chloramphenicol. Using micropipettes 0, 10, 20, 40, 60, 80, and 100 μl (0·0-0·5 ng) of the final standard folic acid solution were added to assay tubes in quadruplicate. The tubes containing standards and sera were incubated for 42 hours. The optical density was read using a Vitatron automated digital colorimeter.

### ROUTINE AUTOCLAVING METHOD

The routine autoclaving method (Waters and Mollin, 1961) was used for comparison.

## Results

Using the 'chloramphenicol' method serum folate levels estimated in 80 blood donors ranged from 2·5 to 14·0 ng/ml with a mean of 6·3 ng/ml. Fourteen

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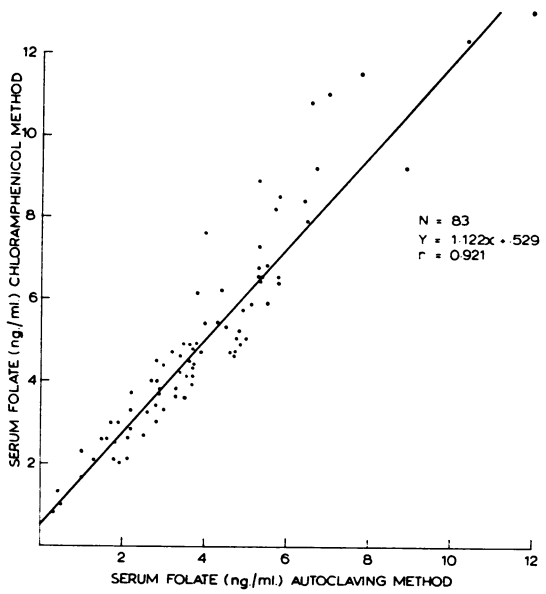


Fig. *Regression analysis comparing 'chloramphenicol' with routine autoclaving methods.*

low serum folate levels in the megaloblastic range (0·3 to 1·3 ng/ml) obtained by the routine method were repeated using the 'chloramphenicol' method. The repeated results ranged from 1·0 to 2·2 ng/ml with a mean of 1·4 ng/ml.

The mean result obtained from the 'chloramphenicol' method was approximately 20% higher than that from the routine method. This difference was revealed when a comparison was made of the results of 83 sera assayed by the 'chloramphenicol' method which does not require autoclaving and the routine method which includes autoclaving (Fig.). The mean correlation (r = 0·921) between the two methods was good and the displacement of the regression line ($y = 1{\cdot}122\,x + 0{\cdot}53$) was in keeping with the difference between the means.

## Comment

The 'chloramphenicol' method presented in this communication is a simple, swift, and economical method for estimating serum folates. The principle is to add micro-quantities of serum and standards directly to medium without aseptic precautions. This is made possible by the use of a chloramphenicol-resistant test organism and micropipettes. These modifications make dilution of serum unnecessary and also obviate the need to autoclave and centrifuge during the assay procedure.

Results obtained from the 'chloramphenicol' technique reveal a 20% higher mean result when compared with those from the routine method. Herbert (1966) also observed higher results using his 'aseptic' method. It would seem that either autoclaving destroys folate despite the presence of ascorbate or that it precipitates protein-bound folate which is available to the test organism when the 'chloramphenicol' or 'aseptic' techniques are used.

We wish to thank the Medical Research Council of Ireland for providing technical assistance in support of this work.

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# Book reviews

**Artificial Organs and Cardiopulmonary Support Systems** By Felix T. Rapaport and John P. Merrill. (Pp. vi + 186; illustrated. $15.00). New York and London: Grune and Stratton. 1972.

This book is reprinted from *Transplantation Proceedings* and covers a symposium on the use of artificial support systems, particularly with a view to holding patients for transplantation. It does concentrate on cardiac support and the problems of long-term extracorporeal bypass and artificial hearts, but artificial respiration and a number of unrelated support systems are also briefly covered. The present state of artificial heart and lung support is extensively covered, and for those concerned in the problems there is useful up-to-date material. Many of the conclusions reached will probably be soon outdated so that this is unlikely to remain a useful reference book for more than a very few years in a developing speciality.

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**Thromboembolism: Diagnosis and Treatment** The proceedings of a symposium held at King's College Hospital, London, sponsored by Kabi Pharmaceuticals Limited. Edited by V. V. Kakker and A. J. Jouhar. (Pp. xii + 241; illustrated. £3·50.) Edinburgh and London: Churchill Livingstone. 1972.

This book contains an assortment of articles from a Symposium on thromboembolism held in July 1971 with the addition of some extracts from the Discussion. It has all the advantages and