

were then injected intraperitoneally into further mice; none of them became infected.

When compared with penicillin, terramycin was found to be more than forty times as potent, weight for weight, because a single subcutaneous dose of 1,000 mgm./kgm. of water-soluble penicillin (calcium salt) did not prevent the relapse of the infection after eleven days.

Single doses of 200 mgm./kgm. of terramycin by mouth prevented the death of mice during the initial parasitaemia, but did not prevent relapse; five daily oral doses of 200 mgm./kgm. were curative, no spiro-nemata appearing in the blood during an observation period of six weeks.

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### A Rapid Turbidimetric Method for the Assay of Antibiotics

RAPIDITY in the assay of antibiotics confers advantages in the control of production, in therapy, as Schneierson<sup>1</sup> has pointed out with respect to aureomycin, and in the requirement of but limited aseptic precautions.

The method described here enables assays to be made with an incubation time of 30 min. The use of a culture of test organism (*Streptococcus agalactiae*) in its logarithmic phase of growth makes this possible. By subculturing twice daily, a rapidly growing strain is maintained; by limiting the incubation period during the night, a vigorous culture is obtained in the morning. (The incubation period may be limited in various ways, such as by the use of a water-bath with a time switch, or by immersing the culture in about two litres of cold water in an insulated flask in an incubator.) From the vigorous culture so obtained, one or two litres of a culture in its logarithmic phase can be prepared in one to two hours. A smaller volume of culture can be maintained in its logarithmic phase by hourly dilution for use at any time during the day. The diluting may be controlled by Spekker readings of turbidity, after a few trials have indicated the turbidity at which the growth-rate begins to diminish; but in practice it soon becomes possible to dilute in a few seconds by eye. In a culture of *Str. agalactiae* in its logarithmic phase, the turbidity increases two- to three-fold in 30 min. Using a 4-cm. cell and starting with a culture giving a Spekker reading on the logarithmic scale of about 0.2 for the difference between culture and clear broth, it is easy to measure the increase due to half an hour's growth with sufficient accuracy.

Serial dilutions of the antibiotic are prepared in sterile broth by adding, with a Record syringe,  $x$  ml. of broth to  $y$  ml. of antibiotic solution, withdrawing  $x$  ml. of this first dilution into a boiling-tube with the same syringe, adding  $x$  ml. of fresh broth to the remaining  $y$  ml. of first dilution with the same syringe, and repeating the operation until enough dilutions have been made. Normally,  $x = y = 5$  for unknown solutions, and  $2x = y = 10$  for standard solutions. By using stops on the stem of the plunger, it is possible to retain solution  $y$  in the barrel of the syringe, the limited movement of the plunger adding,

with adequate mixing, and then subtracting, the volume  $x$  ml. With this technique and a spring-activated plunger, it is possible to prepare sets of serial dilutions with seven dilutions in each set at the rate of one set per minute.

A hand-operated ampoule-filling machine (A. J. Manning, Wembley) is used for adding rapidly and with sufficient mixing 15 ml. of logarithmic culture of test organism to each dilution of antibiotic. After thirty minutes incubation, 1 ml. of 0.02 per cent 'merthiolate' solution (E. Lilly, Basingstoke) is added as violently as possible to each tube, again using a spring-operated syringe. Readings of turbidities on the Spekker followed by the usual graphical comparison between standard and unknown solution enable the concentration of the latter to be calculated.

STANDARD DEVIATIONS OF RAPID ASSAY UNDER VARIOUS CONDITIONS

Anti-biotic	No. of different samples	No. of replicates from each sample	Range of variates	Standard deviation as percentage of general mean	Notes
'Nisin'	25	2	2.9-7.2	9.0	Duplicates assayed on different days
'Nisin'	12	2	1.8-7.7	5.9	Allowance made for differences in 'slope' and for time between duplicates
'Nisin'	9	2	4.1-7.8	3.6	
Penicillin	1	10	—	2.2	One hour's incubation

The method has proved satisfactory during eighteen months use in assaying all kinds of 'nisin' preparations. Preliminary experiments indicate that it might well be successful also with penicillin, streptomycin, aureomycin and gramicidin. It has not been tried with other antibiotics. The accompanying table shows the standard deviations which have been obtained under various conditions.

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<sup>1</sup> Schneierson, S. S., *Proc. Soc. Exp. Biol. Med.*, **74**, 106 (1950).

### Lipoid Bodies, Golgi Apparatus and Zymogen Formation

THE lipid globules (lipochondria) of the Golgi zone of the mouse pancreas have been identified as the Golgi bodies of the live cells by Hirsch<sup>1</sup>, Ries<sup>2</sup>, and Worley<sup>3</sup>. They were, however, studied with the aid of vital dyes or after fixation, and the view that they are vesiculated in life has been open to doubt (cf. Palade and Claude<sup>4</sup>).

Observations on mouse pancreases, depleted of mature zymogen by injections of pilocarpine, have been made on both fresh slices of tissue in 0.85 per cent sodium chloride, and in the pancreas with intact circulation after the method of Hirsch<sup>5</sup>. It has been found that whereas some of the lipochondria are, in fact, homogeneous and not vesiculated, the majority are vesiculated in life. They appear as circles or crescents in cross-section. Though in both the full