

Eine grössere Anzahl Trennblätter von 0,2 mm und 0,3 mm Dicke bieten grösste Variationsmöglichkeiten.

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Summary

A simple tissue-slicer is described. It consists of 10 razorblades mounted in a handle. Details are given how to obtain, with one cut, 9 very similar tissue slices.

PRO EXPERIMENTIS

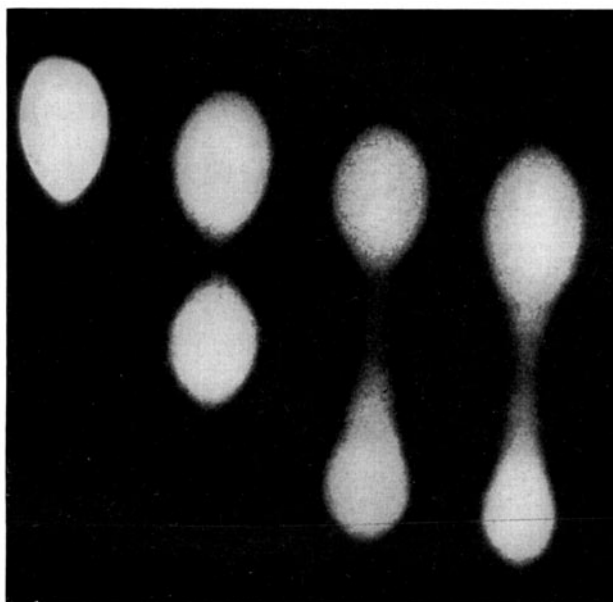
A New Method for Visualizing and Recording Photographically Bioautograms

Working with bioautograms of different forms of cobalamins, especially in liver extracts, obtained by a modification of KOCHER's method¹ (agar plates with *Escherichia coli* 113/3, paper strips applied on the surface for exactly 3 min), difficulties were encountered in the visual observation and photographic record of the zones of exhibition. Looking for a method that did not require the addition of any substance (such as 2:3:5-triphenyl tetrazolium chloride and related compounds)² to the agar before incubation, a number of reactions used to demonstrate peroxidatic activity were tried.

Positive results were obtained, with the appearance of coloured zones corresponding to the sites of growth of *E. coli* 113/3, by pouring on the surface of the agar plates after incubation one of the following reagents: pyrogallol, *p*-phenylenediamine · HCl (both 5% w/v in distilled water), hydroquinone (5% w/v in NaOH 0.1 N), and a mixture 1:1 of α -naphthol and dimethyl-*p*-phenylenediamine · HCl (both 1% w/v), followed by water rinsing and the addition of hydrogen peroxide (1% w/v).

The best results were obtained with *p*-phenylenediamine · HCl. 5 g of *p*-phenylenediamine · HCl are dissolved in 100 ml of distilled water, a little charcoal is added and the solution passed through filter paper. The clear solution is immediately poured on the surface of the agar plate (about 50–60 ml for a plate 31 × 22 cm): after 3 min it is washed away with two-three changes of

distilled water and 50–60 ml of hydrogen peroxide (1% w/v) are added.



Photostatic copy of the paper chromatograms of cyanocobalamin, and three different natural liver extracts (from left to right). Development of the bioautogram with *p*-phenylenediamine · HCl.

In 2–3 min well defined zones with a violet-black colour appear. The hydrogen peroxide solution is washed away and the agar surface is two–three times rinsed with distilled water. Photographic records have to be taken in a few minutes, as a dark background colour soon develops which makes a sharp distinction of the zones of exhibition difficult.

With hydroquinone solutions, used in a similar way, although the zones of growth show a lighter brownish colour, the background does not disturb the photographic records for a few hours.

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Riassunto

Viene rapidamente descritto un nuovo metodo per lo sviluppo e la fotografia di bioautogrammi su piastra d'agar con *Escherichia coli* 113/3.

¹ V. KOCHER, Int. Z. Vitaminf. 26, 321 (1956).

² J. E. FORD and E. S. HOLDSWORTH, Biochem. J. 53, xxii (1953).