v/v). Before the irrigation was completed, about 15 c.c. of crude extract of anthocyanins, for example, from *Viola tricolor maxima*, was pipetted into the column, and chromatographed with half a litre of the organic phase of the solvent mixture.

By this procedure, the anthocyanin mixture was separated into a well-developed chromatogram, as shown in Fig. 1. Then the column was pushed out and cut into bands, and the anthocyanins were eluted with methanol. The methanolic solutions were concentrated, and twice as much benzene was added to obtain an aqueous anthocyanin solution or the crude crystals. They were chromatographed again by the same procedure. In this way, very pure anthocyanins were obtained; for example, violanin and keracyanin from *Viola tricolor maxima*[4]; four other anthocyanins from this plant were not determined on account of their small volume.

Unfortunately, a new batch of Whatman *B* cellulose powder made in 1956 was not so effective. Though the cause is obscure, it is assumed that the separating power of cellulose powders in partition chromatography is conditioned in the same way as that of the adsorbents in adsorption chromatography.

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Dialysis of Small Specimens in Batches

In experiments involving the separation of proteins, it is often necessary to dialyse large numbers of samples, each of a small volume. (Salt fractionation of enzymes, when the fractions cannot be assayed for enzymatic activity in presence of the salt, is a case in point.) Dialysis becomes tedious if large numbers of 1- or 2-ml. samples have to be transferred to dialysis bags from the centrifuge tubes used for precipitating and centrifuging down the fractions, and then have to be recovered more or less quantitatively.

The simple apparatus illustrated in Fig. 1 has proved satisfactory. The rotor consists of a funnel-shaped test tube rack made of polyethylene, with radial ridges which hold the tubes between them. According to the size of the rotor 12–24 tubes can be accommodated at a time.

The tubes containing the samples are covered at the mouths with dialysing membranes, which are secured with small rubber bands. The tubes are checked for leakage by chilling them for a moment, when the membranes should at once contract. The tubes are then inserted between the ridges of the rotor and kept in place by two large rubber bands surrounding the rotor which rest in deep grooves in the ridges. The rotor is then attached by a stainless steel rod to a slow-geared motor and inserted into the bath as shown in Fig. 1. Each tube will then turn along a conical path and thus perform a combined revolving and shaking movement. A rate of 15–25 rotations per min. has been found to provide efficient stirring without causing much foaming. The apparatus also allows dialysis of suspensions, as

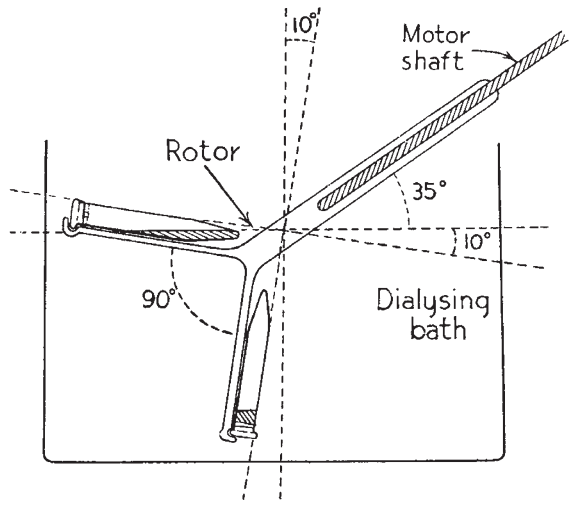

Fig. 1

clogging of the membrane by accumulating particles is avoided.

Using this apparatus, the dialysis of a 1-ml. specimen from a 15-ml. centrifuge tube takes about two to three times as long as dialysing from a bag by the usual method. This disadvantage is, however, for most purposes more than outweighed by the simplified manipulation.

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L Forms of Bacteria and their Role in the Origin of Penicillin Resistance

Investigation of the penicillin resistance of the secondary rods regenerated within the complete or incomplete *L*-cycle is of a great importance for understanding the biological significance of *L* forms of bacteria. So far, only a few authors have occupied themselves with this problem[1–5]. There is one group of authors who maintain that the secondary rods are more resistant to penicillin than the primary ones[5]; on the other hand, there are far more workers who are of the exactly opposite opinion[1–3]. In 1954, we carried out preliminary experiments with *Proteus vulgaris*[6]. It appeared that the secondary rods do not differ essentially from the primary ones with regard to penicillin resistance. These results were confirmed by the following experiments: (1) determination of resistance of the secondary rods regenerated from one large body; (2) determination of the amount of penicillin in broth culture in the time of regeneration of the bacillary form.

(1) Large bodies were isolated from the broth culture containing 1,000 units of penicillin/ml. by the Lindner drop method and the regeneration of the rods was observed in the phase contrast microscope in the moist chamber. The resistance of these rods to penicillin was established by the dilution method in the Lahelle modification[7]. The results were read after 5–7 days and the resistance averaged between