fication and characterization of the separated isoenzymes in analytical gels have, however, stimulated interest in the possibility of obtaining corresponding resolutions on a preparative scale. Several types of apparatus for preparative gel electrophoresis have been described (e.g. Jovin, Chrambach & Naughton, 1964; Gordon & Louis, 1967) and that used in the present study has certain features in common with them in making use of downward migration of protein zones in a vertical, annular gel column with a central condenser for the cooling water and a stream of elution buffer flowing across the lower face of the gel. The elution chamber is of a new pattern, however, which is designed to provide complete elution of protein zones entering the chamber in a small volume of buffer, thus preserving the high resolution of the separation process during collection. The performance of this apparatus has been assessed by the fractionation of alkaline phosphatases from several human tissues and of lactate dehydrogenases from rat tissues in 7.5% polyacrylamide gel in tris-borate buffer, pH 8.6.

For both types of enzyme recoveries of nearly 100% of the added activity were obtained. The relative proportions of the lactate dehydrogenase isoenzymes of rat heart extract determined by preparative electrophoresis were in close agreement with those derived by Wieland & Pfeleiderer (1961) from ion-exchange chromatograms. By contrast, staining and scanning the zones after separation on analytical agar gel electrophoresis underestimated isoenzyme 1. Human small-intestinal and liver alkaline phosphatases were separated by preparative gel electrophoresis, and the heterogeneity within the zones of each enzyme that had previously been demonstrated on analytical gels was preserved in the fractions collected from the preparative column. The closely migrating bone and liver phosphatase isoenzymes could not be resolved completely, but the heterogeneity of a mixture of the two isoenzymes was demonstrated.

The advantages of the apparatus in studies of isoenzymes are thus in providing quantitative recoveries of enzyme fractions with a degree of resolution comparable with that of analytical gel electrophoresis.

The apparatus (patent applied for) is obtainable from Wright Scientific Ltd., 3 Lower Road, Kenley, Surrey.

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The Stoichiometry of Electron Transfer by Bacterial and Plant Ferredoxins

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The number of electrons accepted by spinach ferredoxin on reduction by illuminated chloroplasts was determined by Whatley, Tagawa & Arnon (1963) and by Horio & San Pietro (1964) by measuring the stoichiometry of the reoxidation of the ferredoxin by NADP⁺. Both groups found that spinach ferredoxin accepted one electron/molecule on reduction. Sobel & Lovenberg (1966) measured the number of electrons accepted by bacterial ferredoxin from *Clostridium pasteurianum* by using the method of Whatley *et al.* (1963) and several other methods. They found that clostridial ferredoxin accepted two electrons/molecule on reduction. Tagawa & Arnon (1968) redetermined the number of electrons accepted by clostridial ferredoxin by using the stoichiometry of ferredoxin reoxidation by NADP⁺ or benzyl viologen, and obtained a value for one electron/molecule of ferredoxin.

In the course of our work on the physical properties of ferredoxins we have determined the number of electrons accepted by two bacterial ferredoxins and two plant ferredoxins. The bacterial ferredoxins were from the photosynthetic bacterium *Chromatium* and from the non-photosynthetic anaerobe *Clostridium welchii*. The plant ferredoxins were from spinach and from the blue-green alga *Anacystis nidulans*. By both the methods described by Tagawa & Arnon (1968), we find that both bacterial ferredoxins accept two electrons/molecule on reduction, and that spinach and *Anacystis* ferredoxins each accept one electron/molecule.

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Flavoprotein Properties of D-2-Hydroxyacid Dehydrogenase Purified from Rabbit Kidney

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D-2-Hydroxyacid dehydrogenase (EC 1.1.99.6) is a widely distributed enzyme which catalyses the oxidation of the D-stereoisomer of a number of 2-hydroxyacids to the corresponding keto-acids, using a variety of artificial acceptors (Tubbs &