

long-lived ones will possibly reach a higher enrichment through a more complete soil-plant-soil cycle.

Autoradiographic exposures of plants, plant parts, and of ashed-plant samples demonstrate the same distribution of radioactivity as mentioned above. There is a marked blackening of the photographic film by needles, leaves, twigs, and bark, contrary to the wood which does not affect the photographic film. The autoradiographs show, furthermore, that there are two kinds of radioactive influence. The first and general type causes a slight blackening of the photographic film. The second type is found when examining more recent samples, and it causes the development of small, intensely black dots on the film. These autoradiographic analyses signify that there are at present two sizes of fall-out particles, one very fine-grained and the other more coarse-grained, the latter representing a highly concentrated radioactivity.

PONTUS LJUNGGREN

Geological Institute,
Lund.
March 10.

¹ Ljunggren, P., *Nature*, **184**, 912 (1959).

² Gorham, E., *Nature*, **181**, 1523 (1958); *Can. J. Bot.*, **37**, 327 (1959).

Calcium Binding in Homogenates of Ehrlich Ascites Tumour Cells

WE have recently been attempting to determine the amount of bound (non-diffusible) calcium in the cells of the mouse Ehrlich ascites tumour. Results not yet published indicate that more than 50 per cent of calcium-45 incorporated *in vivo* by the cells is removed by briefly washing them three times in calcium-free balanced salt solution.

In order to investigate further the bound calcium, we disrupted the cells and examined the homogenate. This was done by suspending the cells, which had been labelled *in vivo* with calcium-45, in ice-cooled buffered isotonic salt solution, and then disrupting them by exposure to ultrasonic vibrations of about 20 kc./s. for 2 min. with the *M.S.E.*-Mullard apparatus. The resulting homogenate was subjected to a conventional dialysis procedure at 4-6°C. against a calcium-free solution. Initially calcium-45 was lost from the homogenate very slowly, as indicated by the slight increase in the radioactivity

of the solution against which it was dialysed (Fig. 1). This phase lasted about 90 min., after which calcium was lost rapidly, as shown by the steep portion of the curve of radioactivity of the dialysing solution. A similar curve is obtained even if the isotope is added to homogenized cells *in vitro*. A totally different curve is obtained on dialysing ascitic fluid, in which there is a bound, but exchangeable, calcium fraction. In this case, the increase in radioactivity of the solution against which the labelled fluid is dialysed shows a steep rise from zero time, and flattens only when equilibrium is nearly attained (Fig. 1).

Preliminary experiments also indicate that the total amount of calcium-45 removed from the labelled homogenate by dialysis, even after 4 hr., is considerably less than that extracted by quickly washing the cells in calcium-free solution, and probably amounts to less than 20 per cent of that initially present.

This increase of binding of calcium-45, and hence of total calcium, is corroborated by the very slow migration of the tag towards the cathode from the point of application of the labelled homogenate, when this is subjected to paper electrophoresis.

It appears that, as a result of the disintegration, a number of new calcium-binding sites have been made available. The increase in base-binding capacity may be accounted for, *inter alia*, by (a) molecular damage produced by subjection to the ultrasonic vibrations, or (b) by existing groups, previously inaccessible to the base, being made available by disruption of the cells.

Clearly, a study of the normal distribution of calcium in different cell fractions would be falsified by the increase in the proportion of bound calcium produced by ultrasonic disruption. We are therefore investigating further the nature of the binding in whole cells and in homogenates.

R. SCHOFIELD
D. THOMASON

Christie Hospital and Holt Radium Institute,
Withington, Manchester 20.

VIROLOGY

Purification and Properties of Cauliflower Mosaic Virus

STRAINS of cauliflower mosaic virus occur very commonly throughout North America and Europe. Previous attempts to determine the morphology of the virus¹, and to obtain an antiserum to it², were unsuccessful. Following the recent purification of cucumber mosaic virus³, an attempt was made to purify cauliflower mosaic virus using similar procedures.

The cabbage virus *B* strain of the virus, described by Walker *et al.*⁴, was mechanically inoculated to leaves of young tendergreen mustard plants (*Brassica perviridis* Bailey). Two to three weeks after inoculation, systemically infected leaves were harvested and homogenized in a Waring blender in 0.5 *M* potassium phosphate buffer (pH 7.5) containing 0.1 per cent thioglycolic acid, at the rate of 12 ml. of buffer for each 10 gm. of leaf tissue. The homogenate was strained through cheese-cloth and *n*-butanol added dropwise with constant stirring to give a final butanol concentration of 8 per cent. The mixture then was stirred for 1 hr., after which it was

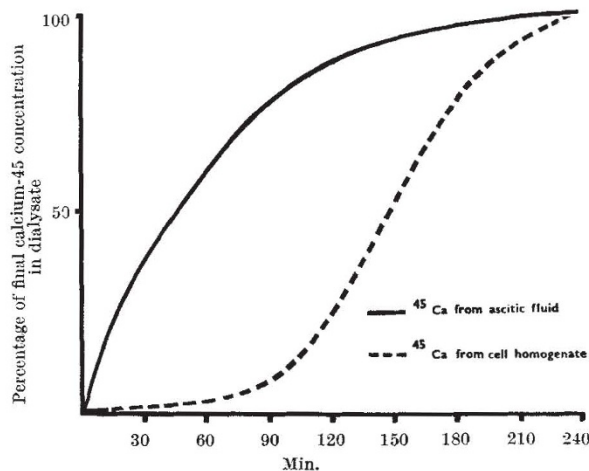


Fig. 1