

METHODS

A METHOD OF CULTIVATING TISSUES IN VIVO IN DIFFUSION CHAMBERS

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A method of constructing diffusion chambers for tissue cultures in vivo described in this paper has many advantages over other methods: reliable airtightness of the chambers, their small size enabling several chambers to be grafted at once in a small animal, absence of formation of a thick connective-tissue capsule around the chambers.

Nowadays the method of tissue culture in vivo in isolated systems based on the use of small chambers made of materials of animal origin or artificial membranes of corresponding porosity, is widely used at the present time in various branches of biological research [1,2].

In this paper, a method of cultivating lymphoid tissue in diffusion chambers differing from those described previously is reported.

Diffusion chambers are made from VUFS brand membrane filters with a pore diameter of 0.1-0.3 μ . To rule out the possibility of toxicity of the material from which these filters are made, tissue cultures are set up in vitro using pieces of filters instead of cover slips. After sterilization in 70° alcohol for 1.5 h, the filter is washed in several portions of physiological saline, and then dried in a sterile Petri dish. To preserve the elasticity of the filter, it must not be completely dried. Under sterile conditions a piece of filter measuring 1 × 2 cm is cut out and folded in half. The size and shape of the rectangles of filter can vary depending on the size of the animals used (mice and rabbits), and if it is necessary to implant several chambers at the same time. A piece of tissue or suspension of cells is placed on the inner surface of the sheet and the edges of the two halves of the filter are glued together. The unfixed edges of the chamber are held together between the blades of a small pair of forceps, leaving 2-2.5 mm free for application of glue made by dissolving pieces of filter in acetone. A completely airtight chamber is obtained, the internal volume of which is quite adequate for growth of the tissue in it, and the method of making the chamber is less laborious than that previously suggested [3]. It must be emphasized that when such chambers are used, the connective-tissue capsules characteristic of chambers assembled on organic glass rings are not observed to form [1,2]. After a suitable time, the chambers are removed from the animal, the glued edges are cut off, and the sheet of filter is straightened out along the line of folding. The total preparation thus obtained is fixed and stained.

The advantages of this make of construction of diffusion chambers are as follows: 1) simplicity of the technique of making the changes and their more reliable airtightness; 2) the possibility of implanting several chambers at once into the same animal and also of grafting chambers into localized areas without injury to surrounding tissues (for example, beneath the capsule of an organ); 3) no thick connective-tissue capsule, which may evidently influence changes in the internal environment and affect growth of the tissue in the chamber, is formed around these chambers.

LITERATURE CITED

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