labeled with TMRITC at a dye-to-protein ratio of 1:40 in a staining period of 4 hr in two different fluorescent-antibody systems. The optimal labeling ratio and staining time for rhodamine-labeled antibody may vary with other immunological systems.

NEW PRESS FOR DISRUPTION OF MICROORGANISMS

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Among a wide variety of methods we have tested for the preparation of cell-free extracts from microorganisms we have had best results, particularly with yeasts, using the Hughes Press (Hughes, Brit. J. Exptl. Pathol. **32**:97, 1951). This apparatus, however, suffers from two major disadvantages: (i) the stainless steel blocks are often difficult to separate after use, and (ii) recovery of the crushed material is tedious and entails considerable loss. The press described here overcomes these disadvantages. The blocks are easily separated at any time during the process, and the crushed, frozen, cell mass is collected almost quantitatively in a plastic centrifuge tube which can readily be removed.

Construction. The cylinder block, bottom plate, and receiving tube block are machined from a cylindrical bar of 303M stainless steel as described in Fig. 1. In addition to the centrally drilled hole, 2 cm in diameter, four equally spaced holes are drilled in one end of the cylinder block and tapped to receive the bolts which attach the bottom plate. The bottom plate has a 1-mm hole drilled centrally, holes for the passage of the four attaching bolts, and two holes drilled partially through to receive aligning pins that are inserted in the top of the receiving-tube block. The receiving-tube block shown has been drilled to accept a 12-ml plastic centrifuge tube. The dotted lines on either side of this hole (Fig. 1) and crossing the bottom of the block represent narrow grooves which allow passage of air around the tube when the press is in operation. The piston is machined from 440C stainless steel, hardened and ground to a diameter 0.05 mm less than the diameter of the cylinder. A shallow mark is cut near the top of the piston to indicate its bottom position in the cylinder. In earlier attempts, pistons made of the same material as the cylinder block were found to be unsuitable because of compression and binding to the cylinder wall under pressure. In an effort to avoid this binding and to assist in aligning the piston in the cylinder, these pistons were tapered slightly from the mid-point toward the top (exaggerated in the diagram) and the top of the piston was rounded. Hardened stainless-steel pistons were subsequently made in this way, although neither modification appears to be necessary, since straight, hardened, stainless-steel pistons with flat tops have been used successfully. While this manuscript was in preparation, details of a similar apparatus were reported (Edebo, Acta Pathol. Microbiol. Scand. **52**:300, 1961). The press described here, however, appears to have certain advantages in simplicity of construction and operation.

Operation. The cylinder block (with bottom plate attached), receiving-tube block and piston are packed in crushed dry ice for a period of at least 20 to 30 min before use. The receiving tube is inserted, the two blocks are assembled, and the suspension of cells to be crushed is run into the cylinder and allowed to freeze. The piston is inserted, and pressure up to about 8,000 psi is applied with a hydraulic laboratory press. When the plunger is fully depressed, the apparatus is removed from the hydraulic press, the cylinder block is lifted off, and the receiving tube removed. Depression of the piston often occurs in short "jerks" and requires from 2 to 15 min. The only difficulty thus far encountered is that with samples of more than 3 to 5 ml occasionally the material freezes around the top of the receiving tube. As more material is extruded through the hole in the bottom plate, the frozen block is forced toward the bottom of the receiving tube, creating sufficient air pressure to break the tube. This has been avoided by using a receiving-tube block that will accept a tube of larger diameter.

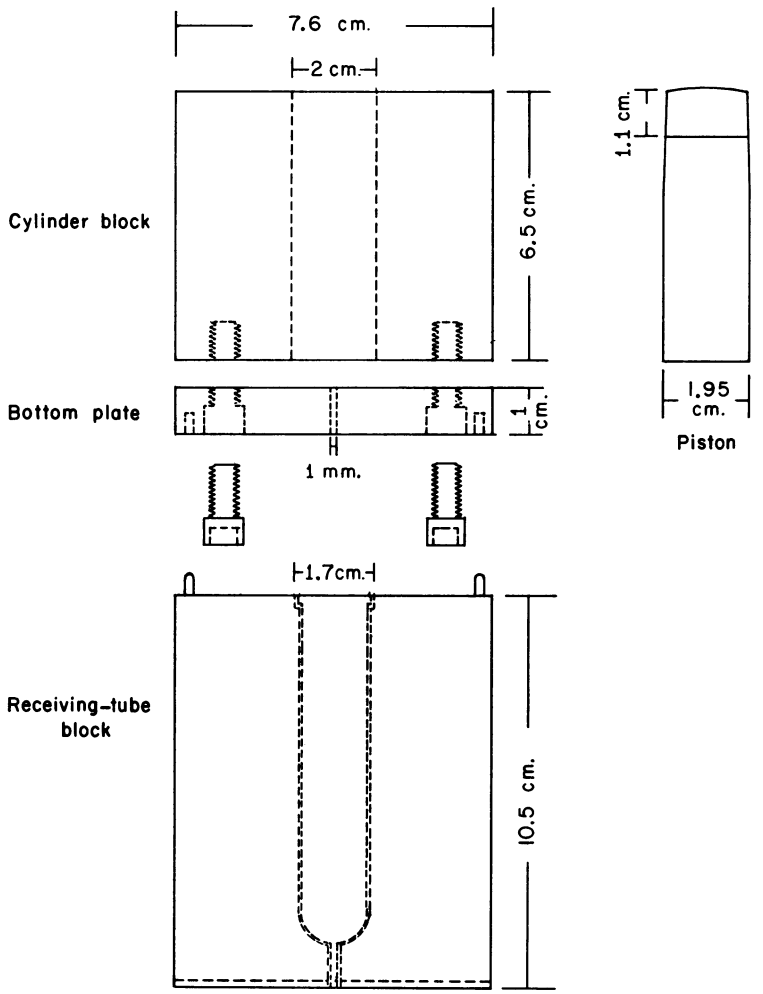

FIG. 1. *Diagram of a new press used for the preparation of cell-free extracts*

The press has been in operation in our laboratory for a period of several months and, although it appears to be equally effective for bacteria, has been used primarily for the disruption of yeast. One strain of *Saccharomyces cerevisiae*, for example, which has been particularly refractory to disruption by other methods, was broken to the extent of from 95 to over 99% in concentrations of 10^8 to 10^{10} cells/ml. Examples of the uses to which the press has been put include the preparation of extracts with high phosphoglucomutase and glucose-6-phosphate dehydrogenase activities, the preparation of particulate fractions with the characteristic 80 S sedimentation constant of yeast ribosomes, and the isolation of yeast deoxyribonucleic acid.

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