Improved Tissue and Cell Homogenizer

CHARLES H. ZIERDT AND GEORGE F. NORRIS

Department of Clinical Pathology, and Biomedical Engineering and Instrumentation Branch, Division of Research Services, National Institutes of Health, Bethesda, Maryland 20014

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Modification of an existing tissue homogenizer is described, which converts to a milk-like consistency even difficult to macerate tissues such as cartilage, hair, and small bones.

Commercially available homogenizers failed to disrupt completely fibrous tissue or bone specimens obtained during autopsy or biopsy procedures in preparation for bacterial culture or enzyme assays.

The conventional cutting blades supplied commercially are all of the general shape seen in Fig. 1. These blades strike the tissue pieces so as to keep them out of the cutting area, where they are exposed only to turbulence.

The glass or Teflon and glass homogenizers of the Ten Broeck type were inadequate for dense tissues, making it necessary to revert to sand, mortar, and pestle to macerate these specimens.

The cutting head on one machine (Omni-Mixer, model 17150, Ivan S. Sorvall, Inc., Newtown, Conn.; similar to Multi-Mix, Lourdes Instrument Corp., Brooklyn, N.Y.) was modified in various ways, and the most effective design, shown in Fig. 1 and 2, was finally adopted.

A stationary multihole collar, to slip over the vertical shaft, was machined from 440-C stainless steel, heat-treated, and electro-polished. The outside diameter of the collar was approximately 1 mm less than the inside of the chamber. It was fastened flush with the end of the shaft with one or two Allen screws, bearing on the vertical shaft. The rotating cutting head was machined to bear against the flat-ground bottom of the stationary collar, providing a positive, scissors-like cutting action. The center hole was threaded to fit the already threaded shaft end. Contact pressure between the rotating cutter and the collar is adjusted by loosening the collar, pressing it against the cutter, then retightening the collar on the shaft. Separate cutting heads are required: one for the microchamber (up to 5 ml), one for the 50-ml chamber, and one for both the 200- and 400-ml chambers. As seen in Fig. 2, the cutting head shaft for the microchamber required addition of an outer supporting shaft for attachment of the fixed, upper half of the cutting head. This supporting shaft is an integral part of the lower screw cover.

Homogenizing action of the new cutter was tested with various tissues from the mouse: muscle, skin and hair, long bones, tails, whole limbs, and blood. HeLa cell, Staphylococcus aureus, and Escherichia coli suspensions were also tested. Saline diluent was used throughout. The cutter and chamber assemblies had been autoclave-sterilized.

Large tissue pieces were cut into smaller pieces of a few millimeters in size before homogenization. Usually 30 sec of full-speed operation was sufficient for complete homogenization. Cooling of the chamber in ice water was utilized for longer periods of cutting action.
No decrease in bacterial colony counts was found after 5 min of exposure, compared to untreated controls, indicating that no bacterial cell disruption occurred. Red blood cells required 5 min for breakage of 90% of the cells, but leukocytes were completely homogenized after 1 min at maximum speed. The rupture of red blood cells was perhaps due more to turbulence than to cutting or grinding action. HeLa cell disruption required 5 min for 75% rupture, as judged by vital staining and counting in a Levy counting chamber.

Long bones from the mouse were rapidly (30 sec) converted to fragments of microscopic size, but reduction of all of these to particles under 50 μm required 10 to 20 min of homogenization. Hair was of similar difficulty to bone, with rapid initial reduction in size, and approximately 20 min of extended treatment was required for further size reduction.

A homogenizer equipped with the new cutter assembly has been completely satisfactory in service for 1 year in the large diagnostic bacteriology laboratory of this hospital, where it has been used to prepare tissues taken at autopsy for bacteriological examination. No maintenance or replacement has been necessary. It has also been used in research applications to disrupt animal tissues for enzyme extraction and for quantitative bacteriological analysis.