2010 GENO/GRINDER®

Tissue Homogenization Cell Lysis

APPLICATION NOTE

Mechanical Disruption for High-Throughput Fatty Acid Extraction from Animal Tissue Samples

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APPLICATION NOTE SP001:
Cell Disruption

APPARATUS:
Geno/Grinder®

APPLICATION:
Cell Lysis

The mechanical homogenization for extraction of fatty acids is traditionally performed with an Ultra-Turrax in a buffer solution. However, both the purification and the slow sample preparation process in the analytical laboratory are labour-intensive. This method is no longer suitable for high sample throughput. Therefore it makes sense to combine the increased sample throughput with mechanical cell disruption. The use of 96-well micro titer plates and deep well plates is suitable for this process. The research in biotechnology/ gene technology also often demands the handling of a considerable number of samples. Sample preparation is often the bottleneck in the process of efficient analysis work. Up to now efforts to mechanically disrupt cells through grinding have been based on the modification of traditional ball or swing mills with an adapter for micro titer plates. The Geno/Grinder is a purpose-built ball mill that was originally conceived for the cell disruption of plant seeds but can also be used for animal and human samples.

Materials and Methods

Two deep well plates (PP), used for collecting and storing of biological materials with a 1.5 – 2ml wells were placed next to each other in the Geno/Grinder clamp assembly. Freeze dried rat muscle or livers were placed in each plate well and spiked with 100mM KH_2PO_4 buffer at pH 2 or trichloro-acetic acid. Then two grinding balls were added. When all wells were filled, the plate was covered with a sealing mat. Thus prepared, the deep-well plates were clamped into the Geno/Grinder. The samples were shaken vigorously for one minute at 1700 strokes per minute. This resulted in a fine, homogeneous sample dispersion within each well. Because of the vertical movement of the Geno/Grinder clamps, all wells are moved uniformly. This results in uniform and reproducible homogenization yields for all samples. As well as using titer plates with 96 wells it is also possible to use other formats, e.g. the "Biomek micro centrifuge 24 position tube rack" for Eppendorf Safe-Lock reaction tubes.

Results and Discussions

The results show that the Geno/Grinder is well suited for cell disruption of muscle and internal organs, i.e. liver. However, the quality of the results depends on the grinding accessories. Using several small Zirconium oxide beads with a diameter of 2mm does not give a satisfactory result. The sheer force is not sufficient to grind frozen muscle tissue. In contrast to this, it was possible to successfully homogenize the frozen material with stainless steel grinding balls of 4mm diameter.

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