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# BONE SLICES



**Bone slices are for accurate *in vitro* assessment of osteoclastic bone resorption either by traditional staining or by use of the CrossLaps® for Culture ELISA assay.**

Product number #1BON1000

**LITERATURE:** 1. HENRIKSEN ET AL. AM J PATHOL 164:1537-1545 (2004). 2. HOLLBERG ET AL. EXP CELL RES 279:227-238 (2002). 3. IVASKA ET AL. J BIOL CHEM 30:279(18):18361-9 (2004). 4. KARSDAL ET AL. AM J PATHOL 166:467-476 (2005). 5. SCHALLER ET AL. J BONE MINER RES 19:1144-1153 (2004). 6. STROUP ET AL. J BONE MINER RES 16:1739-1746 (2001).

ISO 9001 certified

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# BONE SLICES

Bone is a dynamic tissue that is remodelled through life. It is believed that special factors are present in bone, which may affect the activity of osteoclast. Thus at Nordic Bioscience a/s we use high quality bone, not dentine, for all our research. We now provide this service for our customers.

Each batch of bone slices is tested for quality by resorption assays for verification of osteoclastic ability to make pits on these exact bone slices. Subsequently, the supernatant is measured by the Crosslaps® for Culture ELISA kit and only if both tests are positive the bone slices are accepted for further resorption assays.

The ability of osteoclast to make pits is tested by the human osteoclastic resorption assay (Karsdal et al. J Biol Chem 2003). A correlation of quantification of osteoclastic resorption either by the traditional staining of resorption pits or the Crosslaps® for Culture ELISA assay is shown to the right in figure 1 and 2 (Schaller et al. J Bone Miner Res 2004).

Since osteoclast experiments often demands higher amount of protein that can be extracted from bovine bone slices used in 96 well plates, Nordic Bioscience a/s have developed cortical bone slices that fits perfectly into either 6, 12, 24 or 48 well plates, as illustrated in figure 3.

Human osteoclasts were differentiated and plated on bovine cortical slices. The antiresorptive compound was added in different concentrations (A: 90 µM, B: 30 µM, C: 10 µM, D: 0 µM) for 6 days. On day 6 an aliquot of the cell culture supernatant was used to determine the CTX amount (CrossLaps® for Culture ELISA) and the osteoclasts were removed from the bone slices and stained with myosin hematoxylin to visualize the pit formation (figure 1).

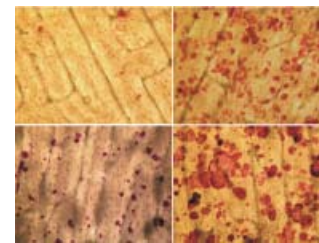
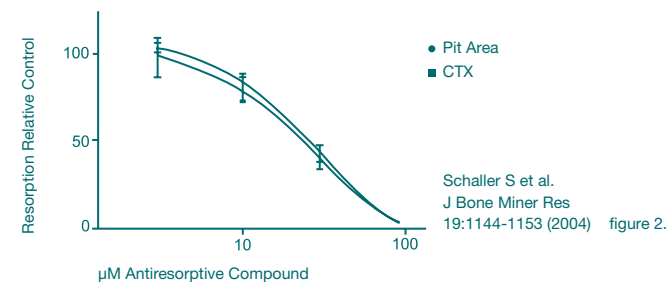


figure 1.



The concentration of the compound (log) was plotted against the CTX value to give (or produce) a dose response curve (figure 2).

Nordic Bioscience a/s is using these large bone slices for 6 well plates (Giant Bone Slices (GBS)) for protein extraction of osteoclasts cultured on bone instead of plastic, which have shown to influence on protein expression (data not shown). We recommend using these substrates for e.g. RNA extraction for gene profiling experiments for osteoclasts. Both protocols for RNA isolation from bone slices and culture protocols for human osteoclasts can be obtained upon request.

## ORIGIN

The bone slices are made from the cortical part of the femur of bovine bones, and is perfectly fitted for the 96 well format. The bone slices are 6 mm in diameter and approx. 200 µm thick.

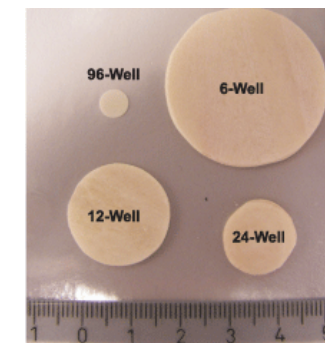


figure 3.

## STORAGE

The bone slices are stored in 70% ethanol at 4°C. However, for shorter time periods and during transport room temperature is adequate for retaining the quality of the bone slices.

## HANDLING

We recommend that our bone slices are washed 3 times in 10% serum containing medium before they are transferred to the 96 well plates.

## CULTURE

Usually resorption experiments lasts 72 hours (as presented in figure 2), however already after 16 or 24 hours after addition compound to osteoclast cultures a significant differences are observed. Even after 8 hours a significant difference can be observed in optimised assays.

## FEATURES

In contrast to pit staining and scoring the use of Crosslaps® for Culture ELISA does not require termination of the osteoclast culture. Thus, several samplings can be obtained from the same bone slice, allowing for e.g. reversibility measurement of osteoclast activity in response to given compounds.

The material is disposed of as normal cell culture material.