



HOMOGENIZATION AND EXTRACTION OF FLUORESCENT CHEMICAL COMPOUNDS FROM MOUSE BONE TISSUE

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/ CONTEXT

Atlanthera is a drug discovery company specialized in bone-targeted drug delivery through covalent binding of drugs to hydroxybisphosphonates (HBP), a class of molecule known for high affinity to bone. Extraction of HBP-conjugated chemical from bone is challenging and require efficient tissue homogenization. Precellys® Evolution system was tested for bone homogenization and compared with a cryogenic grinder.

/ MATERIALS

- · Instrument: Precellys® Evolution
- Lysing kit: Hard tissue grinding MK28-R reinforced 2mL tubes with 2.8mm stainless beads (KT03961-1-008.2)
- Tissue samples: mouse tibia (50 to 110 mg)
- · Homogenization buffer: PBS
- Extraction buffer: 6N HCl, CHCl₃/methanol
- Suspension buffer: acetonitrile

/ RESULTS

The best homogenization efficiency was obtained at 8500 rpm on dry rather than homogenization in buffer.

Compound extraction was equivalent to our standard protocol (cryogenic grinder)

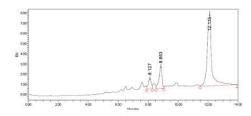


Figure 1: Example for HPLC analysis of compound extracted from bone tissue after homogenization with Precellys* Evolution

/ PROTOCOL

 $Precellys^{\$}$ Evolution, 8500 rpm, 8x10 sec (10 sec breaks), room temperature

Samples: Mouse tibia were removed, cleaned from soft tissues, snap frozen in liquid nitrogen and stored at -20°C. Frozen tibia were either dry processed or processed in PBS buffer. Homogenized tissue was suspended in extraction buffer. Organic phase was purified, evaporated and the dry extract was resuspended in acetonitrile prior to HPLC analysis.

/ CUSTOMER



/ CONCLUSION

Precellys® Evolution associated to the appropriate lysing kit is a suitable, simple and convenient homogenization system to extract chemical compounds from mouse bone tissue. Compared to our standard method (cryogenic grinder), Precellys® Evolution is much faster (shorter homogenization time, less manipulation steps and larger number of sample per cycle) and is free of cross-contamination between samples thanks to its single use consumables.

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