



RatLaps™ ELISA

Bone Resorption Marker (CTX-I) For Monitoring Preclinical Trials

Assessment of bone resorption in rodents

- Ovariectomized (OVX) rats and mice
- Thyroidparathyroidectomized (TPTX) rats
- Rat and mouse models of bone metastases
- Knock-out and transgenic mice with effect in bone resorption

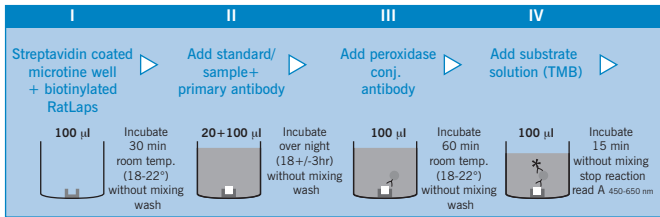
Assessment of bone resorption in tissue culture

- Calvariae, tibiae, or metatarsals from rat and mice

The RatLaps™ ELISA is used for quantitative assessment of bone resorption in rodent bones. The assay detects C-telopeptide fragments of collagen type I (CTX-I) generated during osteoclastic bone resorption in rat and mouse urine, serum and cell culture supernatant.

RatLaps™ ELISA

ENZYME IMMUNOASSAY FOR QUANTITATIVE ASSESSMENT OF BONE DEGRADATION IN RODENTS



Performance Characteristics

Method:	• Competitive ELISA
Format:	• 96-well microplate with reagents sufficient to test 40 samples in duplicate
Detection limit:	• 2.0 ng/ml
Analyte:	• Sequence (EKSQDGGR) specific for a part of the C-terminal telopeptide $\alpha 1$ chain of type I collagen (CTX)
Specimen:	• Serum or plasma, urine, cell culture supernatant
Specimen volume:	• 20 μ l
Precision CV intraassay:	• < 10%
Precision CV interassay:	• < 15%
Species Reactivity:	• Rat, mouse, guinea pig
Shelf life:	• 6 month
Assay time:	• Overnight

Sampling

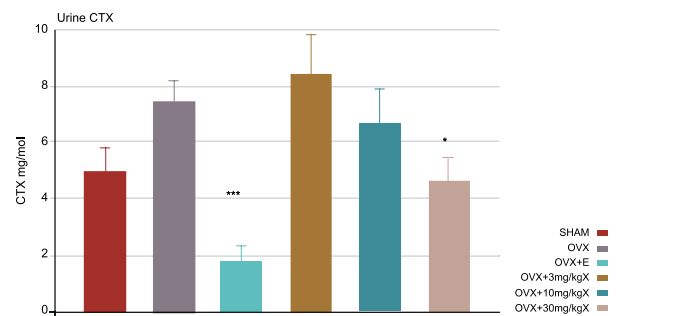
Collection of blood or urine samples after overnight fasting period. Collect either spot urine or 24-hour urine. Freeze samples for prolonged storage (<-18 °C).

Expected serum values (females)		mean \pm SD
SPRD	3 month	38.8 \pm 6,6 ng/ml
	6 month	26,9 \pm 7,0 ng/ml

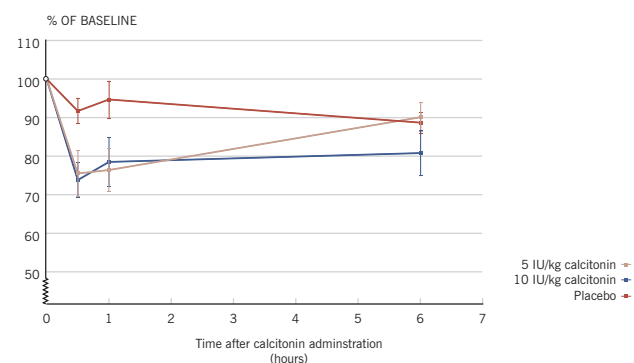
The RatLaps™ ELISA kit for research use only.
Product number # 1RTL4000

Assay Procedure

1. Add 100 μ L of **Biotinylated RatLaps Antigen** to each well, cover with sealing tape, and incubate for 30 \pm 5 minutes at room temperature (18-22°C).
2. Wash the immunostrips 5 times with **Washing Solution**.
3. Add 20 μ L of **Standards, Control**, or samples into the appropriate wells followed by 100 μ L of **Primary Antibody**. Cover the immunostrips with sealing tape and incubate over night (18 \pm 3 hours) at 2-8°C. Wash as in 2.
4. Add 100 μ L of the **Peroxidase-conjugated Antibody** to each well, cover with sealing tape, and incubate for 60 \pm 5 minutes at room temperature (18-22°C). Wash as in 2.
5. Pipette 100 μ L of the **Substrate Solution** into each well and incubate for 15 \pm 2 minutes at room temperature in darkness.
6. Pipette 100 μ L of the **Stopping Solution** into each well.
7. Measure the absorbance at 450 nm with 650 nm as reference within 2 hours.



Urine CTX level was determined as a biochemical parameter of bone resorption. Six-months old OVX rats were treated orally with the indicated doses (mg/kg) of compound NS3736, 17-b-estradiol (E), or vehicle (V) once daily for 6 weeks or were sham operated. ***p<, significant different compared with OVX vehicle control; *p < 0.05, significant different compared with 3 mg/kg; #p=0,08 significant different compared with OVX vehicle control.



RatLaps™ assess the effect of anti-resorptive drugs in vivo rapidly

LITERATURE: 1. GARNERO ET AL. J BONE MINER RES 18:859-867 (2003). 2. HOEGH-ANDERSEN ET AL. ARTHRITIS RES THER 6:R169-R180 (2004). 3. HOSEA ET AL. EXP BIOL MED (MAYWOOD) 229:303-311 (2004). 4. ISHIKAWA ET AL. J RHEUMATOL 31:1174-1179 (2004). 5. KATSUMATA ET AL. MAGNES RES 18:91-96 (2005). 6. KRUGER ET AL. BR J NUTR 94:244-252 (2005). 7. LEFORT ET AL. UROL INT 74:301-307 (2005). 8. PENNISI ET AL. J BONE MINER METAB 23:134-139 (2005). 9. SCHALLER ET AL. J BONE MINER RES 9:1144-1153 (2004). 10. TIVESTEN ET AL. J BONE MINER RES 19:1833-1839 (2004).

all the way

FROM RESEARCH TO PATIENT MONITORING

Nordic Bioscience Diagnostics A/S • Herlev Hovedgade 207 • 2730 Herlev • Denmark • www.nbdiagnostics.com