

CALCEIN STAINING

Protocol used for calcein bulk staining of microcracks

Created by Silvia Owusu (✉ silvia.owusu@zmk.unibe.ch) on September 29 2021
Created by Mathieu Simon (✉ mathieu.simon@artorg.unibe.ch) on September 29 2021

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Protocol purpose:

This protocol is aimed to stain in vivo microcracks by performing bulk staining of 3 mm thick bone samples using calcein. Calcein binds to free calcium present at the surface of microcracks and bulk staining allow to stain microcracks present in vivo only, not the ones generated by sample preparation.

PROTOCOL

Step 1 [~? + ? minutes]

Cutting

- Cut bone sample using a butcher saw into slices of 3 mm thickness.

Step 2 [~? + ? minutes]

Fixation

- Do something

Step 3 [~? + ? minutes]

Bulk staining

- Dissolve calcein at a concentration of 0.5 mM. Dissolution is performed either in distilled water or in 100% ethanol.
- Perform bulk staining under low vacuum (~300 mbar) at room temperature. 2 samples are stained for 7 hour and 2 are for 24 hours.

Step 4 [~? + ? minutes]

Embedding

- Do something

Step 5 [~? + ? minutes]

Sample preparation

- Do something

Step 6 [~? + ? minutes]

Histology analysis

- Do something

WARNINGS



Calcein

The $C_{30}H_{26}N_2O_{13}$ is bad for you. In case of contact, wash directly and inform the safety responsible



Human tissue

Human bone can transmit diseases. Always use gloves when manipulating

NOTES

REFERENCES