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| Título | Decalcification with nitric acid and EDTA may influence on Safranin O staining: a study in different rodent strains |
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Justification: The decalcification process for microscopic analysis of mineralized samples in pathology is a challenge. Whereas the quality of results of the histochemical staining is related to the choice of decalcifying agent. Several times decalcification agents affect morphology and cause damage to proteins which may compromise the results of laboratory analyzes. Aim: The objective of this study is to evaluate the effects of two types of descaling agents on the special staining of safranin O in the different rodent strains (AG/WT, BALB/c, C57, DBA1/J mice and Wistar rats). Materials and Methods: Our study was approved by the Animal Ethics Committee of Hospital de Clínicas de Porto Alegre (HCPA) (no. 17-0381) and conducted in accordance with National Institutes of Health (Bethesda, MD) guidelines. The right and left hind paws were removed and placed in 10% buffered formalin. The specimens of each lineage were randomly divided into the following three groups: (1) 10% nitric acid (n=7), (2) 12.5% EDTA (ethylenediamine tetra-acetic) at room temperature (n=7), and (3) 12.5% EDTA at 35°C and shaking (n=7). Joint sections were obtained by microtome and we evaluate the intensity of matrix stained in red using Safranin O staining. The intensity score was assessed and the staining was classified as: absent, weak, or strong. Results: Due to the low number of samples (animals per protocol per lineage), there was not a significant statistical difference. The results show that in Wistar strains, EDTA did not show absent coloration in the samples, but nitric acid also had a strong staining index. In AG/WT, BALB/C, and C57 strains, no staining absent in EDTA samples was observed, and almost 100% weak or absent staining with the use of nitric acid. In the DBA1/J strain, EDTA showed mostly strong staining, without any weak, and nitric acid a greater proportion of samples that were weak staining. Although there was no statistical difference, in Wistar strains the use of nitric acid does not seem to be so harmful in this type of staining.