

## **Statistical analysis**

Descriptive statistics will be performed for the pre-defined variables. Normality of distribution of residuals and homoskedacity will be checked first for the continuous analyzed data. A fisher's exact test will be performed to assess any differences in the qPCR positive samples between the three interventions in all stages in terms of final irrigation with NaOCl and the two supplemental procedures. Univariable and multivariable (median) regression analysis will be performed to assess the effect of different supplemental procedures on the reduction of bacterial load in teeth with apical periodontitis. Interaction effects will be checked through the likelihood ratio test. Effect sizes and 95% CIs will also be presented. The level of statistical significance will be set at 95%. All analyses will be conducted with Stata version 15.1 (Stata Corporation, College Station, Texas, USA).