

Research paper 7

## The development of a simple protocol for successful inking of surgical margins with easily accessible colouring agents

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**Background:** Inking enables precise assessment of the surgical margins, which has important therapeutic implications.

**Objectives:** To determine the impact of using acetic acid and the method of air drying and fixation on the retention, intensity and the original colour of the agent used.

**Method:** A total of 120 lipomatous tissue sections measuring 1x1x0.5 cm were used, 60 sections were painted with Fevicyl-black and 60 sections with Fevicyl-orange, the best and worst colours as determined in a previous study, respectively. 40/120 sections were used to assess the effect of acetic acid (20 with and 20 without acetic acid), and 20 sections each to assess four different methods of air drying and fixation. All sections were routinely processed and stained. Three scores were obtained by two independent investigators; the retention along the circumference from 1-4 (1-25%-1, 26-50%-2, 51-75%-3, 76%-100%-4), the intensity and preservation of the original colour (each graded as 1 - barely visible at x400, 2 - visible at x400, 3 - visible at x100, 4 - visible at x40). A composite grade was obtained by the sum of the Z-scores of these three. Two-way analysis of variance (ANOVA) and a post hoc mean comparison (Tukey HSD) were performed to analyse the data.

**Results:** There was a significant difference in standardized composite scores with respect to the use of acetic acid in case of the worst colour, orange ( $p = 0.0483$ ) but none when considering the four methods adopted in air drying/ fixation.

**Conclusion:** It is recommended that acetic acid is used routinely when inking specimens to obtain optimal results.

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