EVALUATION OF STAINING TECHNIQUES ON THE RESULTS OF MICRONUCLEUS IN EXFOLIATED ORAL MUCOSA CELLS

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INTRODUCTION

Micronuclei (MN) in exfoliated epithelial cells are widely used as biomarkers of cancer risk in humans *(El-Zein et al., 2006, 2008)*. MN are classified as biomarkers of the breakage and loss of chromosomes *(Mateuca et al., 2006)*. They are small, extra nuclear bodies that arise in dividing cells from centric chromosome/chromatid fragments or whole chromosomes/chromatids that lag behind in anaphase and are not included in the daughter nuclei in telophase *(Zalacain et al., 2005; Fenech et al., 2006; Utani et al., 2007)*. Buccal mucosa cells have been used in biomonitoring exposed populations because these cells are in the direct route of exposure to ingested pollutants, are capable of metabolizing proximate carcinogens to reactive chemicals, and are easily and rapidly collected by brushing the buccal mucosa *(Salama et al., 1999)*.

The **OBJECTIVE** of the present study was to further investigate if, and to what extent, different stains have an effect on the results of micronuclei studies in exfoliated cells. These techniques are: Papanicolaou (PAP), Modified Papanicolaou, May-Grünwald Giemsa (MGG), Giemsa, Harris's Hematoxylin, Feulgen with Fast Green counterstain and Feulgen without counterstain.

METHODOLOGY

Seventy cytology smears (2 per individual) were taken from the buccal mucosa by scraping with endobrush. Ten smears were stained with each technique. The slides were evaluated under a light microscope with 1,000-fold magnification using oil immersion according to the parameters of nuclear, micronuclear, citoplasmatic staining intensity and staining samples. The final score range could be between 0 – 20, whereby a slide is considered to be satisfactory when the final score is at least 12.

RESULTS

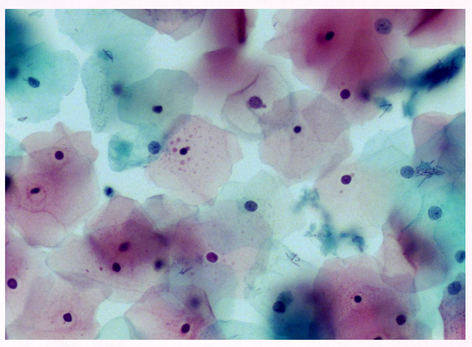

Figure 1 – PAP

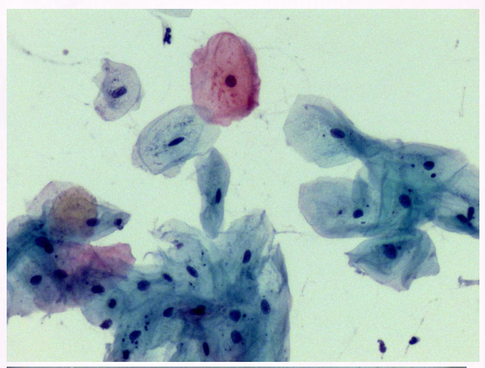

Figure 2 – Modified PAP

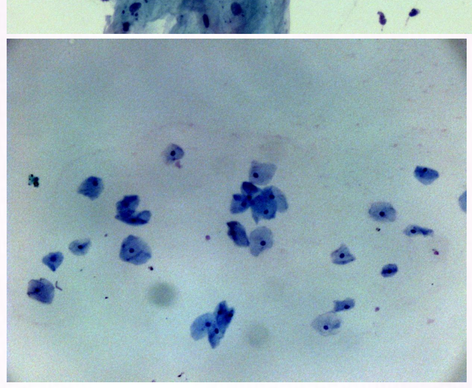

Figure 3 – MGG

Table 1 - Final score for each tecnhique

Technique	Final Score
PAP	14
Modified PAP	14
MGG	12
Giemsa	12
Harris Hematoxylin	11
Feulgen & Fast Green	4
Feulgen without counterstain	20

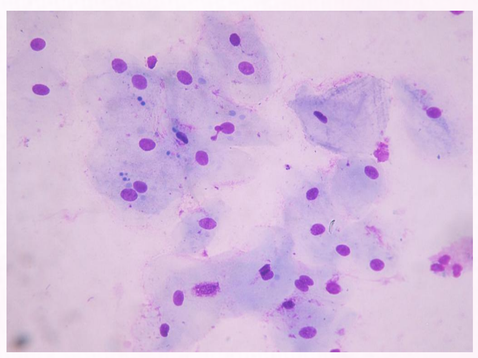

Figure 4 – Giemsa

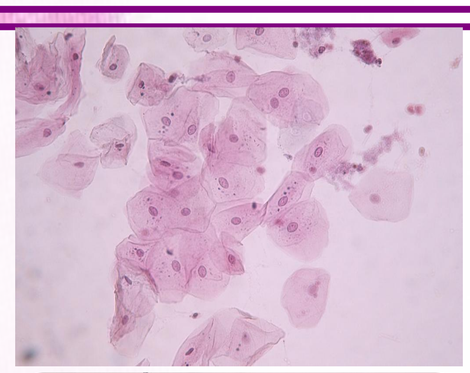

Figure 5 - Hematoxylin

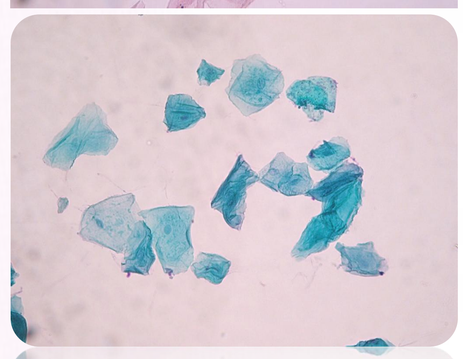

Figure 6 - Feukgen & Light Green

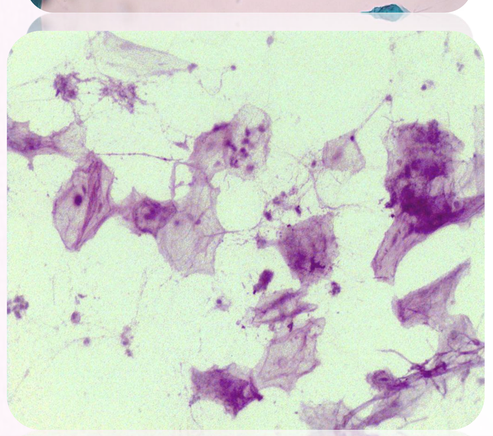

Figure 7 - Feulgen without counterstain

CONCLUSIONS

The results with Feulgen with Light Green as counterstain and Harris's Haematoxylin were not satisfactory. All other results were classified as satisfactory, with PAP and Modified PAP stains scoring 14 and MGG and Giemsa scoring 12 . The higher and maximum score (20) was obtained with Feulgen without counterstain.

Feulgen without counterstain was the preferred method for detecting MN in buccal mucosa cells due to its specificity to DNA like the study performed by *Nersesyan et al., 2006*.

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