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STUDIES IN ORAL LEUKOPLAKIAS

IV. MITOTIC ACTIVITY IN ORAL LEUKOPLAKIAS

A PRELIMINARY REPORT

by

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Observations from the Department of Oral Pathology, Royal Dental College, Copenhagen, associate some oral leukoplakias classified as hyperparakeratosis with epithelial atypia, carcinoma in situ or carcinoma. This association has not been observed in leukoplakias showing solely hyperorthokeratosis. (*Pindborg, Poulsen, Grete Renstrup & Silverman* 1963). Mitotic activity, a possible indicator of malignancy, has not been determined in oral leukoplakias. The purpose of this report is to describe a method of estimating epithelial mitotic activity, and also to compare the findings regarding this activity within the two groups of oral leukoplakic lesions classified histologically as hyperparakeratosis and hyperorthokeratosis.

METHOD AND MATERIAL

The method used in this study for determining mitotic activity is a modification of *Marthaler's* technique (1956) in which the mitotic activity is expressed in terms of number of mitotic figures

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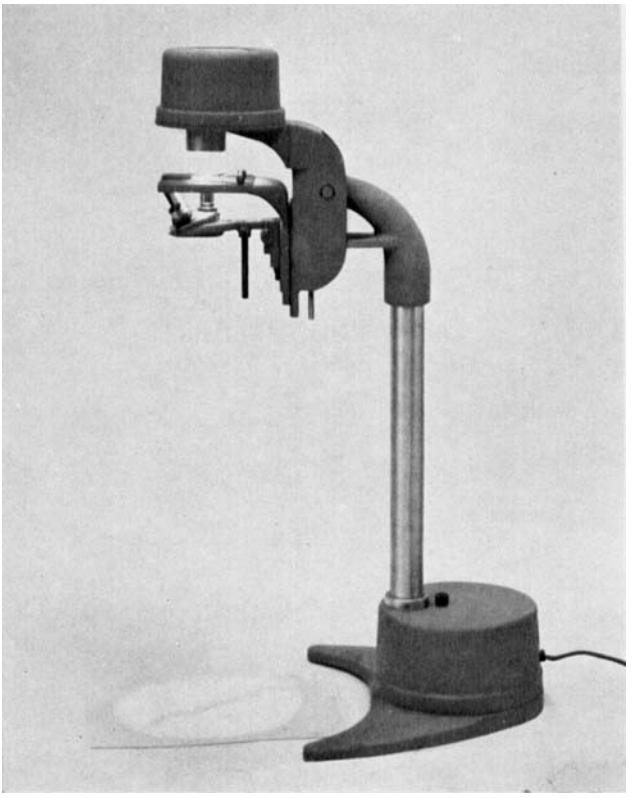


Fig. 1. Projector used for tracing sections.

per unit length of basement membrane. Tracings are made of serial sections magnified to 80 times by use of a projector, Fig. 1. Precautions should be taken that the selected areas are not cut tangentially. Measurements of length are accomplished by superimposing a nylon thread along the basement membrane, Fig. 2. Each section is then placed under the microscope and at a magnificating of about 600 times, all the mitoses (Fig. 3) within an outlined area are counted and marked on a drawing, Fig. 2.

Histologic material for this investigation was obtained from two groups of patients: one group of 10 males, ages ranging from 41 to 72 (average age 62 years), with lesions characterized histo-

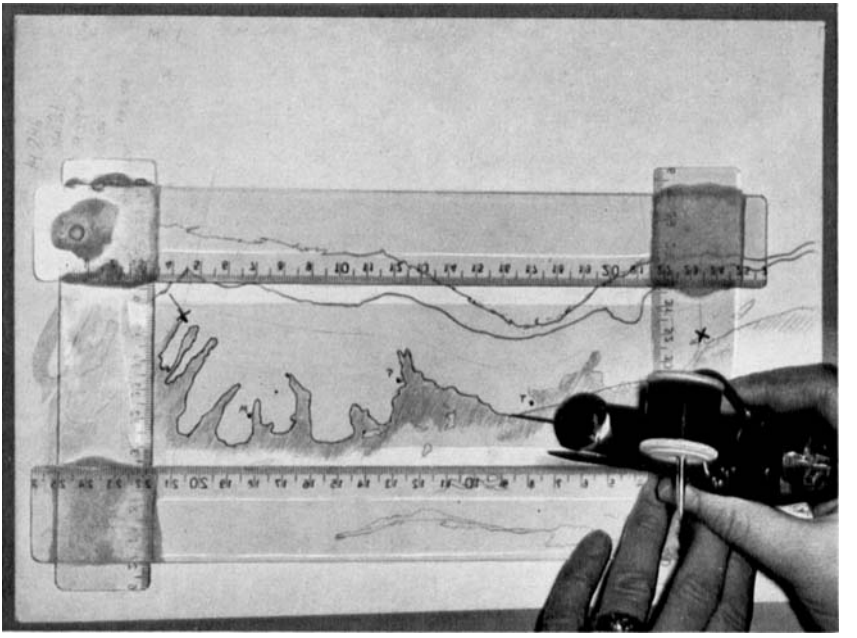


Fig. 2. Drawing of a section with a nylon thread superimposed on the basement membrane. Mitoses are marked.

logically by hyperparakeratosis; the second group of 8 males and 2 females, ages ranging from 38 to 71 (average age 55 years), with lesions histologically classified as hyperorthokeratosis, Fig. 4 A and B.

To obtain as uniform material as possible, only leukoplakic lesions in the buccal mucosa were examined in this study. The biopsies were immediately fixed in 10 % formalin, embedded in paraffin, and serially cut in 50 sections of 6 μ thickness. Every fifth slide was stained with hematoxylin and eosin. Using every fifth section assures a sufficient distance to avoid counting the same mitotic figure twice.

FINDINGS

Fig. 5 illustrates the increased mitotic activity of hyperparakeratotic lesions as compared to those of hyperorthokeratosis.

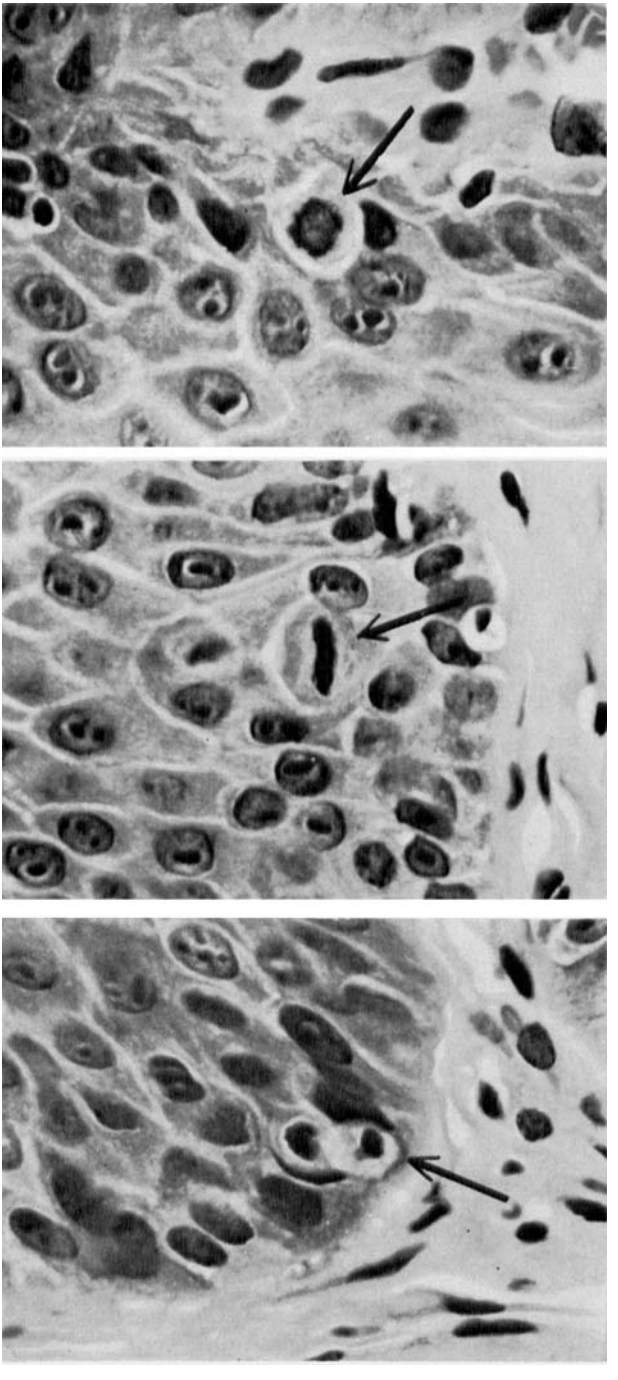
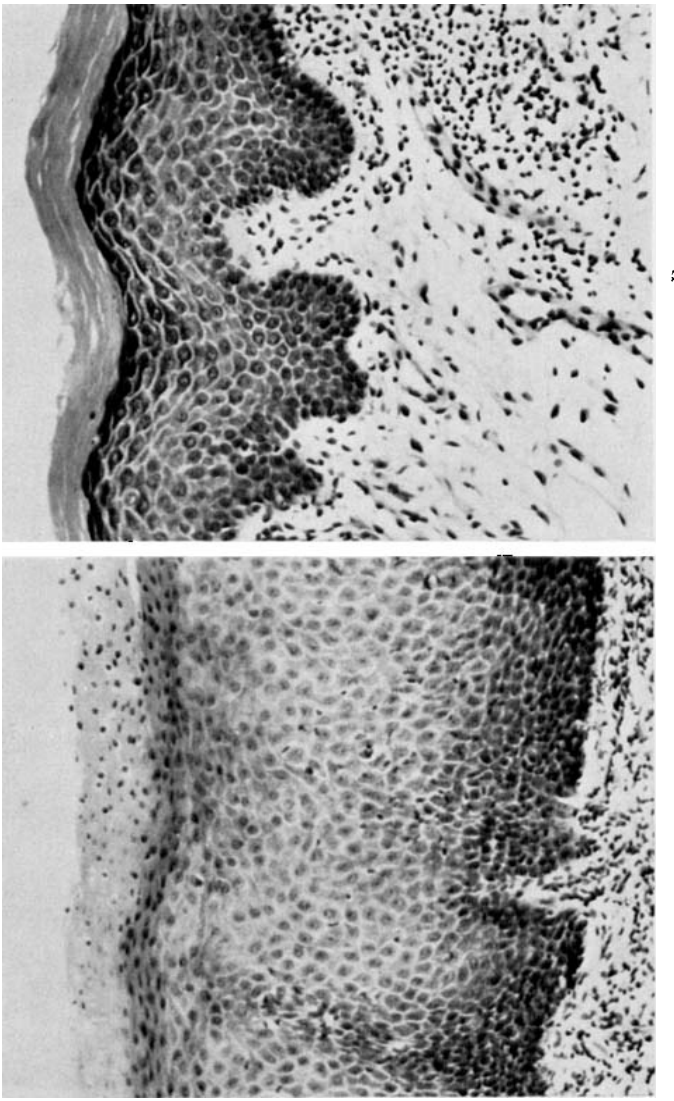


Fig. 3. Examples of various phases of mitoses. Note occurrence of lymphocytes among basal cells in epithelium. Magnification: × 600.



B.

A.

Fig. 4. A. Hyperparakeratosis in section from leukoplakic lesion in buccal mucosa. Magnification: × 150.
B. Hyperorthokeratosis in section from leukoplakic lesion in buccal mucosa. Magnification: × 150.

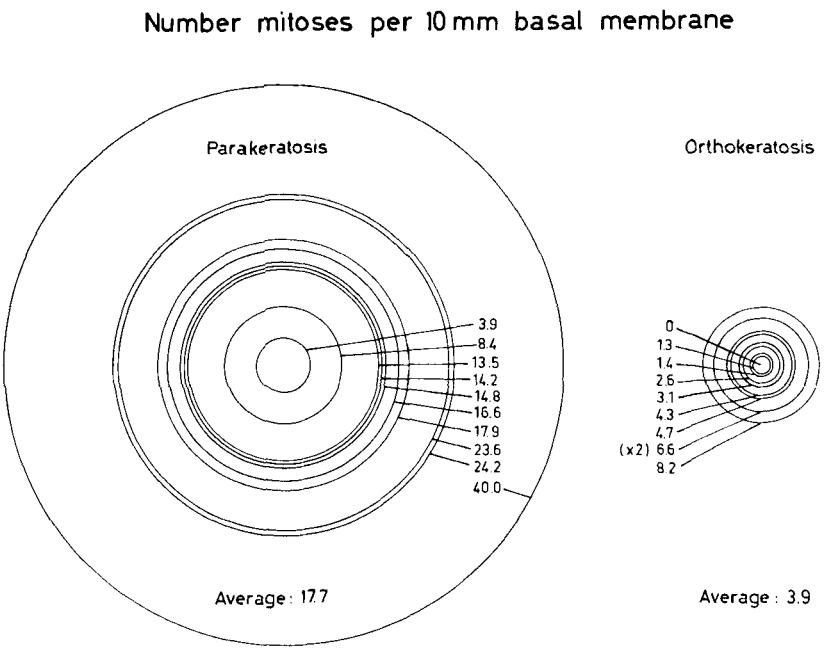


Fig. 5. Graphic demonstration of mitotic activity in a group of 10 hyperparakeratotic lesions and a group of 10 hyperorthokeratotic lesions.

The average mitotic activity of the hyperparakeratotic group was more than 4 times that of the hyperothokeratotic group. There was no apparent correlation with age or sex, although this series is too small to justify any conclusion in this respect. Table 1 illustrates calculations necessary to arrive at a reproducible figure expressing mitotic activity.

DISCUSSION

Previous studies of oral mucosa dealing with mitotic activity have utilized the so-called mitotic index which indicates the number of mitotic figures found per 1,000 counted cells (*Julia Meyer, Marwah & Weinmann* 1956). There are several reasons why this technique was considered inadequate for the present investigation. Oral leukoplakias sometimes exhibit increased density of cells in the basal cell layers often associated with epithelial hyperplasia. Furthermore, heavy infiltration with

Table 1

Example of determination of mitotic activity for one patient with a parakeratotic lesion.

Slide no.	Number of mitoses	Length of basement membrane
1	5	285 mm
6	9	319 „
11	5	341 „
16	9	421 „
21	6	467 „
26	13	554 „
31	16	509 „
36	9	445 „
41	11	463 „
46	13	487 „
Total	96	4291 mm

Magnification: × 80

96 M per $\frac{4291}{80}$ mm basement membrane

17.9 M per 10 mm basement membrane

lymphocytes and plasma cells makes it difficult to distinguish nuclei necessary for cell counting.

This study shows a definite difference in mitotic activity between two small groups of patients with oral leukoplakias classified histologically as either hyperparakeratosis or hyperorthokeratosis. Previous observations indicate an association of some hyperparakeratotic oral leukoplakias with epithelial atypia, carcinoma in situ or carcinoma. Therefore, it might be assumed that there exists a correlation between high mitotic activity and malignant potential.

The mitotic activity expressed in number of mitoses per unit length of basement membrane as described in the present investigation is reproducible. If the mitotic activity is a reflection of malignant potential, this activity could be useful for further classifying oral leukoplakias.

Acknowledgement

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SUMMARY

By means of a new technique for determining mitotic activity in epithelium, two groups of oral leukoplakias were examined and compared. The average mitotic activity of the group histologically characterized by hyperparakeratosis was 4 times higher than that of the group histologically characterized by hyperorthokeratosis.

RÉSUMÉ

ÊTUDES SUR LES LEUCOPLASIES BUCCALES
IV. INDEX MITOTIQUE DANS LES LEUCOPLASIES BUCCALES. RAPPORT PRÉLIMINAIRE.

A l'aide d'une technique nouvelle pour déterminer l'index mitotique dans l'épithélium, deux groupes de leucoplasies buccales ont été étudiés et comparés. L'index mitotique du groupe caractérisé au point de vue histologique par une hyperparakératose était 4 fois plus élevé que celui du groupe caractérisé au point de vue histologique par une hyperorthokératose.

ZUSAMMENFASSUNG

UNTERSUCHUNGEN ÜBER LEUKOPLAKIEN DER MUNDSCHLEIMHAUT
IV. MITOTISCHE AKTIVITÄT BEI LEUKOPLAKIEN DER MUNDHÖHLE. VORLÄUFIGER BERICHT.

Unter Anwendung einer neuen Technik zur Bestimmung der mitotischen Aktivität im Epithel wurden zwei Gruppen oraler Leukoplakien untersucht und mit einander verglichen. Die durchschnittliche mitotische Aktivität in der Gruppe, welche histologisch hyperparakeratotischen Charakter trug, was 4 mal grösser als in der, welche histologisch durch Hyperorthokeratose charakterisiert war.

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