

Sex Determination with Sex Chromatin of Epithelial Cells from the Oral Mucosa in Koreans*

Yung Keun Oh, Tai Sun Shin and Jong Sun Kim

*Departments of Anatomy and Dermatology
Yonsi University College of Medicine*

ABSTRACT

The authors studied 100 Koreans for sex determination using the sex chromatin of the epithelial cells from the oral mucosa according to Marberger's method. Turbid suspensions obtained by scraping the buccal mucosa were used; these were stained with a modified Feulgen technic for DNA.

Summarizing our observations, the percentage of the sex chromatin in the nuclei of the epithelial cells showed a remarkable difference between the male and female of 23.6%. The authors confirmed the fact that the method of oral smear which had already been introduced by Marberger and his associates (1955) might be applicable to the Korean, also.

Sexual dimorphism in the nuclei of several tissues has recently been described by Moore et al. (1953) and by Moore and Graham (1954). Using the skin biopsy technic of Barr, Marberger and Nelson (1954) have studied the genetic sex in cases of pseudohermaphroditism. Polani et al. (1954) and Wilkins et al. (1954) have also studied ovarian genetics.

Thereafter, there has been a natural trend to investigate methods which would avoid the operative procedure required for a skin biopsy. Davidson and Smith (1954) reported on the morphological sex differences in polymorphonuclear neutrophil leucocytes. Marberger et al. (1955) first introduced the oral smear as a method of chromosomal sex determination. Nakadate et al. (1956) also presented statistics on the incidence of sex chromatin in cells from the oral

mucosa.

Based on the reports above the authors have conducted this study in order to determine the genetic sex in cases of pseudohermaphroditism and for medicolegal purposes.

MATERIALS AND METHODS

The authors examined specimens from 100 Koreans (50 males and 50 females). Those with exudative inflammation or with tumors within their buccal cavities were excluded. Turbid suspensions obtained by scraping their oral mucosa with a small metal spatula were used. Immediately after smearing these suspensions on a glass slide, they were fixed in a modified Davidson's solution for 12 to 24 hours and were stained according to a modified Feulgen technic for DNA.

The staining procedure was as follows: for hydrolysis, the materials were placed in normal hydrochloric acid (preheated) at 60°C for 10 minutes and immersed in Schiff reagent for 10 minutes and then bathed in M/20 sodium bisulfate and counterstained with light green (0.5% aqueous light green 5 parts and 80% alcohol 95 parts).

All prepared slides were observed under oil immersion. In making the counts, the authors counted the sex chromatins in 100 intact nuclei from each slide, and adopted a method which Marberger et al. (1955) had established for identifying the sex chromatin. All the sex chromatin which was counted in each slide was classified into three shapes, three positions and two sizes.

RESULTS

The results are shown Table 1. In 50 female cases,

* This investigation was supported by a research fund from the Research Council of Yonsei University College of Medicine. The authors wish to express their gratitude to Dr. K. D. Choi for the privilege of examining his patients and to Dr. S. Y. Pak for his invaluable suggestions.

sex chromatin was found in 7% to 59% of 100 nuclei, an average incidence of 23.7% with a range of 52%. In 50 male cases, sex chromatin was found in 0% to 1% of 100 nuclei, an average incidence of 0.1% with a range of 1%. The sexual difference in incidence of sex chromatin obtained from the epithelial cells of the oral mucosa was 23.6%.

Table 1. Incidence of sex chromatin in cells from the oral mucosa in Koreans

Sex	Age group	No. of cases	Incidence	
			Range, %	Avg., %
Females	Newborns to 15 yr.	31	7-49	20.6
	16 to 50 yr.	19	11-59	26.7
	Total Newborns to 50 yr.	50	7-59	23.7
Males	Newborns to 15 yr.	29	0-1	.1
	16 to 57 yr.	21	0-1	.2
	Total Newborns to 57 yr.	50	0-1	.1

DISCUSSION

As mentioned above, the average incidence sex chromatin was 23.7% in the female. Comparing this result with others, it was found that, in spite of using the same materials, there were great differences between the results. Marberger et al. (1955) reported that the average incidence of sex chromatin in the female is 45.6%, while Nakadate and his associates, figure for this is 19.73%.

The authors considered that these variations might be caused mainly by a difference in standards in identifying sex chromatin. Nakadate et al. included only the typical planoconvex sex chromatin lying adjacent to the nuclear membrane, which was called by them a localized thickening of the nuclear membrane. But Marberger et al. included not only the sex chromatin lying adjacent to the nuclear membrane but also that lying free in the karyoplasm.

The second reason for this difference might be that due to technical differences. Nakadate et al. used an alcohol ether solution (alcohol 50 parts and ether 50 parts) as the fixative and a cresyl echt violet as the stain, while Marberger et al. and the authors used the modified Davidson's solution as the fixative and the modified Feulgen technic as the stain.

Comparing the average incidence in the female with that in the male, it was evident that a remarkably wide difference existed between them. The sexual difference in our work was 23.6%, while Marberger et al. reported this to be 45% and Nakadate et al., 19.35%. However, all reported a remarkable difference between the sexes, although there were great variations in incidence among the authors, and all reported that it would be a reliable standard for determining the genetic sex.

It was interesting to note that the average incidence of sex chromatin obtained from the nuclei of human skin using the skin biopsy method of Moore et al. (1953) showed a sexual difference similar to that in the epithelial cells from the oral mucosa. Moore et al. found sex chromatin in nuclei of cells from the female skin in 52% to 85%, with an average incidence of 69%, and from the male skin in 1% to 14%, with an average incidence of 5%, an average sexual difference of 64%, a difference greater than in the nuclei of cells from the oral mucosa. But the authors could not deny the fact that the skin biopsy method for chromosomal sex determination had the shortcoming of complexity in preparation, though with the advantage of greater certainty in detecting sexual differences.

REFERENCES

- Davidson, W. M. and Smith, D. R.: *Brit. M. J.*, 2:6, 1954.
- Lillie, R. D.: *Histopathologic technique & practical histochemistry*, Blakiston Co., Phila., p. 132, 1954.
- Marberger, E., Boccabella, R. A. and Nelson, W. O.: *Proc. So. Exp. Biol. Med.*, 89:488, 1955.
- Marberger, E. and Nelson, W. O.: *J. Clin. Endocrin. Metab.*, 14:768, 1954.
- Moore, K. L. and Barr, M. L.: *Acta Anat.*, 21:197, 1954.
- Moore, K. L., Graham, M. A. and Barr, M. L.: *Surg. Gynec. Obst.* 96:641, 1953.
- Nakadate, K. H., Yasosima, S. S., Isono, N., Mukai, S. and Takada, D.: *Jap. Med. J.* 1702:14, 1956.
- Polani, P. E., Hunter, W. E. and Lennox, B.: *Lancet* 2:120, 1954.
- Wilkins, L., Grumbach, M. M. and Van Wyk, J. J.: *J. Clin. Endocrin. Metab.* 14:1270, 1954.

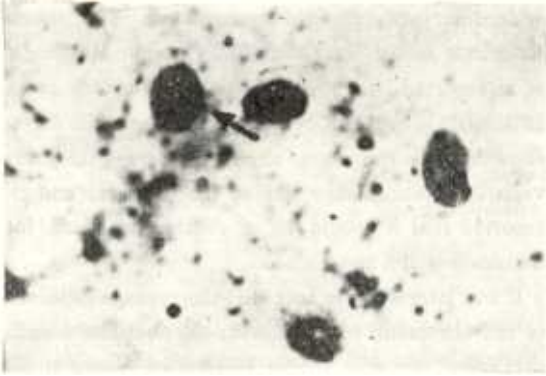


Fig. 1. A typical planoconvex sex chromatin is indicated. Note its characteristic position adjacent to the nuclear membrane. An adult woman, modified Feulgen technic. $\times 1000$.

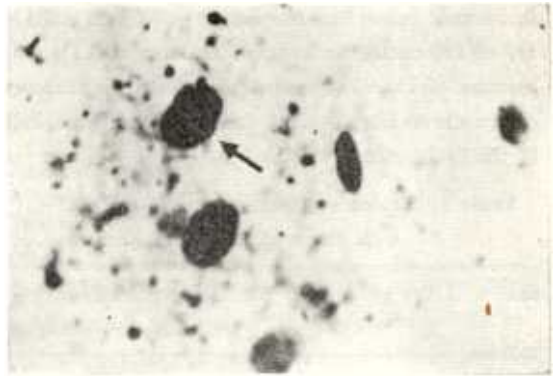


Fig. 3. An ovoid sex chromatin lying free in the karyoplasm is indicated. 6-year-old girl, modified Feulgen technic. $\times 1000$.

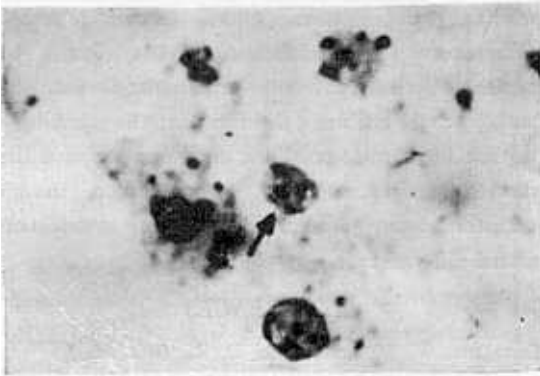


Fig. 2. A round sex chromatin lying adjacent to the nuclear membrane is indicated. An adult woman, modified Feulgen technic. $\times 1000$.



Fig. 4. Typical male nuclei. Note absence of sex chromatin. 12-year-old boy, modified Feulgen technic. $\times 1000$.