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Original Article

Sex Chromatin Positive Cells in the Buccal Smears of Normal Newborn Females

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ABSTRACT

The present study has been conducted on normal 100 newborn females, born at Umaid Hospital, Dr. S. N. Medical College, Jodhpur (Rajasthan). The aim of the study was to find out frequency of sex chromatin positive cells in buccal smear of normal newborn females and their relation with the birth weight. The slides were prepared from the buccal smears and stained by the Carbol Fuschin method. The sex chromatin bodies were present in range of 3-11% with an average of 6.4 ± 0.25%. We found no relation between the number of sex chromatin positive cells and the birth weight of newborn females.

1. Introduction

The sex chromatin is seen in interphase nuclei of human beings and other mammals. It is one X chromosome heavily condensed along its entire length. Cajal [1] was first to describe the presence of paranuclear mass in cats and human beings. Later on it was described by Barr and Bertram [2] in neuron cell of cat and is known as Barr body or Barr corpuscle. Moore and Barr [3] demonstrated the existence of sex chromatin in the cells of mucosa of cheeks of normal human females. The sex chromatin is seen as planoconvex intranuclear chromatin body measuring between 0.7-1.2 microns in size. It is closely applied to the inner aspect of nuclear membrane. It has an affinity for basic dyes and contains DNA, but considerable amount of RNA is also present. It is assumed to be a female sex chromosome as it is not found in normal males. It is visible in the body cells when the embryo is about 2 weeks old.

The sex chromatin has gained much practical value in the medical world. It can be well demonstrated especially in smears of the easily accessible mucous membranes e.g. the buccal smear test or mouth epithelium test for nucleo-morphological determination of sex. The knowledge of sex chromatin has also resulted in advances in various other fields' i.e nuclear morphology and cytochemistry, nucleoprotein synthesis, tissue culture, antenatal sexing of foetuses, tumor including cancer and forensic medicine. Inspite of so much importance being attached to it, very little work has been done in our country about the study of sex chromatin in buccal smears in normal human beings, so this study has been conducted.

2. Material and methods:

The present study has been conducted on normal 100 newborn females, born at Umaid Hospital, Dr. S. N. Medical College, Jodhpur (Rajasthan). Buccal smears were collected on second post natal day from inside of cheek with a wooden spatula. The bacterial flora from the oral cavity can be eliminated by antiseptic mouth washes as it is not possible in newborns so we discarded first smears. Smears were spread over albumenized slides. They were fixed by keeping them in a fixative (absolute alcohol-95 cc and Distilled Water-5 cc) and fixation time was ½-24 hours. Then smears were hydrated using 80%, 70%, 50% alcohol in that descending order and water. The slides were kept for 2-5 minutes in each concentration of alcohol and stained by Carbol Fuchsin Method [4]. Then differentiate them in 95% ethyl alcohol. Put slides in absolute alcohol varying from few dips to 1 minute. Clear them in xylene and absolute alcohol (equal parts).Clear them in xylene. Mount them in canada balsam. Examine slides with binocular microscope under oil immersion lens, count nuclei and record number of nuclei containing sex chromatin body.

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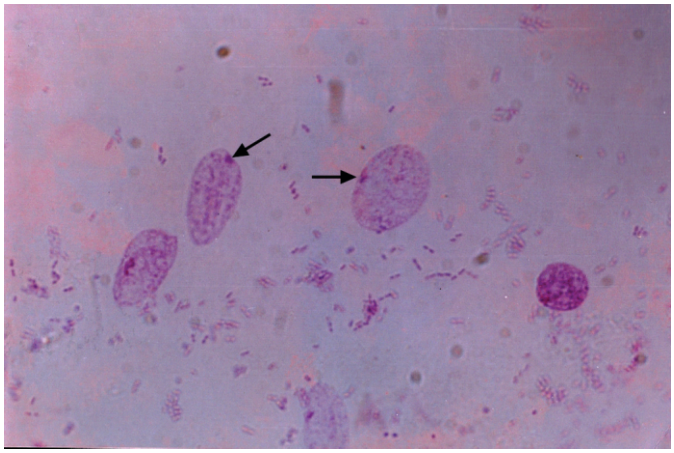
3. RESULTS

Table I shows relation of birth weight to the frequency of sex chromatin positive cell in buccal smears. The weights of newborn ranges between 1500-4500 grams. Maximum cases were between 2500-3000 grams and mean was 6.4 ± 0.25.

Table I: Relation of birth weight to the frequency of sex chromatin positive cell in buccal smears

Weight of Newborns (gms)	No. of Observations	Range	Incidence of Sex Chromatin %	
			Mean	S.E.M.
1501-2000	8	3-8	6.5	±0.21
2001-2500	36	4-10	6.9	±0.35
2501-3000	37	5-11	6.4	±0.32
3001-3500	14	3-7	6.2	±0.34
3501-4000	4	4-9	6.1	±0.31
4001-4500	1	7	6.6	±0.40
Total	100	3-11	6.4	±0.25

Figure I: 1 & 9 O' clock position showing the sex chromatin in buccal smear of newborn females



4. Discussion:

The sex chromatin is not due to XX chromosomes but is due to the fact that one X chromosome is tightly coiled along most or all of its length during interphase. As it is tightly coiled so it become visible when stained. The sex chromatin is not present in the nuclei of males as though they also have one X and Y chromosome because X in the males does not coil but remains uncoiled (extended) in interphase nuclei. This extended chromosome gives information that both X and Y chromosome are derived from autosomes but during evolution Y chromosome has lost most or all of the genes which are not concerned with the determination of sex. The X chromosome has retained about 50 genes which have nothing to do with sex. If these genes which control the information of enzymes do not work, the body cells of male would suffer from many defects. So an active extended X chromosome is essential both in the body cells of males and females. One of the X chromosome remain coiled in female and other remains extended because if both X chromosome are in extended form then enzyme production will be excessive which can cause serious effects.

In our study the incidence of sex chromatin positive cells in the buccal smears of newborn females on second post natal day ranged from 3 to11%. The majority of the observations fell between 4 to 8%. The mean of total 100 observations was 6.4 ± 0.25. The results of present series are in consensus with other series reported by Aggarwal et al, Taylor and Majumdar et al [5, 6, 7]. The incidence of sex chromatin to some extent depends upon the shape of sex chromatin body. If it is plump, the incidence is usually high and if it is flattened against the nuclear membrane then the incidence may be low because of difficulty in recognizing it. The technical factors may also greatly influence sex chromatin percentage in the nuclei. The fixatives vary greatly in their preservation of nuclear details. The formalin may often, but not always produce such homogeneity of the chromatin pattern, that the sex chromatin may be obscured. Other fixatives may also act this way or block the staining of sex chromatin. The long period in strongly acid fixatives may cause nucleotides to hydrolyze out, so that staining characteristics will be poor. If the stain used strongly colors the nucleolus, other cell components or bacteria, the sex chromatin will be difficult to be recognize or may marked entirely. Chapelle [8] believed that everyone who interprets oral smears develop a system of his own. Some discard all nuclei with minor irregularities whereas some use less strict criteria. In oral smears, nuclei of very thick or thin film show low percentage. Forsberg [9] mentioned that experience of investigators and what type of cell are discarded and what cells are inspected will make a difference in results.

Eidenbenz [10] and Homma and Kajii [11] reported higher frequency of sex chromatin in new born with low birth weights whereas Majumdar et al [7] examined the new born who had birth weight of 2000 gm or less, the incidence of sex chromatin was significantly lower than that in other groups. But in our study we found no relation between the number of sex chromatin positive cells and the birth weight of newborn females.

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