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# Chromosomal Abnormalities in Severely Malnourished Indian Children*)

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## ABSTRACT

The frequencies of chromosomal aberrations in Giemsa-banded preparations obtained from peripheral blood cultures of 5 kwashiorkor, 3 marasmic and 2 marasmic-kwashiorkor patients were investigated. Analysis of 1120 metaphases from these patients revealed an aberration rate of 5.80 per cent as against a frequency of 1.66 per cent seen in control children. However, no particular chromosome or chromosome group seems to be more affected in malnourished children. Furthermore, malnourished children with associated infections were observed to exhibit more number aberrations than those without infections. These observations thus, support the suggestion that malnourished children exhibit increased frequencies of chromosomal damage.

## INTRODUCTION

Cytogenetic studies in children sufferring from severe protein energy malnutrition (PEM) have produced controversial results. While some investigators[1,2,8,10,19], have observed increased frequencies of constitutional chromosomal abnormalities in malnourished children, others[3,18,20,21], have failed to confirm these findings. Furthermore, when the results have been positive, the question has been raised of whether they are due to associated factors like vitamin defficiencies, anemia or infections most commonly present in malnourished children.

A carefull "controlled" study has therefore been undertaken to investigate the frequencies of constitutional chromosomal anomalies in severly malnourished children with and without infections and who exhibited no clinical signs of vitamin defficiency or anemia. In addition an attempt has also been made to test whether any specific chromosome or chromosome group is more involved for damage in malnourished children.

## MATERIALS AND METHODS

Ten children sufferring from severe PEM, comprising 5 kwashiorkor, 3 marasmic and 2 mixed type were investigated. Their clinical data are given in Table 1. Four of these children were having either gastrointestinal or respiratory infections at the time of cytogenetic analysis. Also, no patient had either anemia or signs of vitamin defficiency. Ten children apparently disease free, with body-weights ranging between 80-100% of the standard weight/age served as controls[13].

Peripheral blood samples were obtained from the patients and control children after informed consent. A part of the sample was used for measuring serum albumin[6], and haemoglobin levels[22].

Lymphocytes in whole blood were cultured in RPMI-1640 medium supplemented with foetal

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**Table 1.** Cllnical data of malnourished children

| Case No. | Sex | Age (months) | Clinical type of PEM | Weight (g) | Weight for age deficit (%) | Infections* |
|---|---|---|---|---|---|---|
| 1. | M | 12 | Kwashiorkor | 5,000 | 39 | Yes |
| 2. | F | 15 | Kwashiorkor | 5,450 | 37 | No |
| 3. | M | 12 | Kwashiorkor | 5,600 | 41 | Yes |
| 4. | F | 17 | Marasmus | 5,700 | 50 | Yes |
| 5. | F | 18 | Marasmus | 5,900 | 40 | No |
| 6. | F | 20 | Kwashiorkor | 5,950 | 42 | No |
| 7. | M | 20 | Mixed | 6,250 | 37 | No |
| 8. | M | 12 | Kwashiorkor | 4,510 | 45 | No |
| 9. | F | 60 | Mixed | 8,400 | 54 | No |
| 10. | M | 15 | Marasmus | 5,430 | 48 | Yes |

* Include either gastrointestinal or respiratory infections. No patient had a known viral infection or vaccination at the time of study. Also, no patient had been exposed to X-rays before cytogenetic analysis.

calf serum and antibiotics as described previously[12]. Phytohaemagglutinin (Wellcome) was added and the cultures were incubated for 52h. Colchicine was added during the last 1h of incubation and the metaphase preparations were made as described previously[12]. The preparations were stained for G-banding as described[12]. At least 100 consecutive metaphases were scored for each subject. The aberrations were classified into individual chromosome or chromosome group according to the Paris Conference[14].

## RESULTS

Nomal children had a mean level of $3.79 \pm 0.14$ g/dl (Mean $\pm$ SEM) serum albumin as against a value of $2.05 \pm 0.01$ in malnourished children. The mean haemoglobin level in normal and malnourished children were $10.9 \pm 0.08$ g/dl and $11.1 \pm 1.0$ respectively.

Data on the frequencies of chromosomal aberrations are presented in Table 2. A total of 2,320 mitoses were analyzed, 1,120 from malnourished children and 1,200 from controls. Malnourished children exhibited a mean aberration rate of 5.80% in contrast to a rate of 1.66% seen in controls. This difference was statistically significant. The aberration observed in malnourished children comprised chromatid-type and chromosomal-type. However, the chromatid-type aberrations were more commonly seen. Of the aberrations, breaks and gaps constituted a major fraction, followed by dicentrics and fragments. No rings and exchange figures were observed.

**Table 2.** Frequency of chromosome aberrations in normal and malnourished children

| Case No. | No. of metaphases analysed | No. of abnormalities | Per cent abnormalities |
|---|---|---|---|
| *Normals* | | | |
| 1. | 125 | 2.0 | 1.6 |
| 2. | 150 | 4.0 | 2.7 |
| 3. | 100 | 1.0 | 1.0 |
| 4. | 100 | 0.0 | 0.0 |
| 5. | 100 | 0.0 | 0.0 |
| 6. | 150 | 3.0 | 2.0 |
| 7. | 175 | 5.0 | 2.9 |
| 8. | 100 | 1.0 | 1.0 |
| 9. | 100 | 2.0 | 2.0 |
| 10. | 100 | 2.0 | 2.0 |
| Total | 1200 | 20.0 | 1.66 |
| *Malnourished* | | | |
| 1. | 100 | 11.0 | 11.0 |
| 2. | 125 | 8.0 | 6.4 |
| 3. | 100 | 5.0 | 5.0 |
| 4. | 105 | 4 0 | 3.8 |
| 5. | 100 | 9.0 | 9.0 |
| 6. | 110 | 12.0 | 10.9 |
| 7. | 120 | 3.0 | 2.5 |
| 8. | 120 | 7.0 | 5.8 |
| 9. | 135 | 2.0 | 1.4 |
| 10. | 105 | 4.0 | 2.9 |
| Total | 1120 | 65.0 | 5.8* |

* Significantly different from controls ($\chi^2$, $P<0.001$)

**Table 3.** Interchromosomal distribution of chromosome aberrations

| Subjects/ Chromosome or chromosome group | A | | | B | | C+X | D | E | F | G+Y | Total aberration |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | | | | | | |
| Controls | 3 | 3 | 2 | 3 | 1 | 4 | 2 | 2 | — | — | 20 |
| | (15.0) | (15.0) | (10.0) | (15.0) | (5.0) | (20.0) | (10.0) | (10.0) | | | (100.0) |
| Malnourished children | 10 | 11 | 6 | 6 | 8 | 9 | 7 | 4 | 1 | — | 62* |
| | (16.1) | (17.7) | (9.7) | (9.7) | (12.9) | (14.5) | (11.3) | (6.5) | (1.6) | | (100.0) |

Figures in parenthesis indicate percentages. Ten subjects were studied in each group.
* Fragments were excluded.

**Table 4.** Frequency of chromosome aberrations in malnourished children with and without infections

| Group | No. of children | Total no. of metaphases analysed | No. of abnormalities | Per cent abnormalities |
|---|---|---|---|---|
| Children with acute infections. | 4 | 435 | 40 | 9.1 |
| Children without acute infections. | 6 | 685 | 25 | 3.6* |
| ... Total ... | 10 | 1120 | 65 | 5.0 |

* Significantly different from the value of 1.66% seen in control children. ($P<0.01$: where P is by $\chi^2$ test).

Data on the interchromosomal distribution of aberrations are presented in Table 3. The chromosome aberration frequency appears to be a simple function of the size of the chromosome. More number of aberrations were recorded on the longer chromosomes than on the shorter, as expected.

In an attempt to find out the role of infections in the observed incidence of chromosomal aberrations, the results were expressed according to the presence and absence of infections in the malnourished children. The data are presented in Table 4. It can be seen that malnourished children with infections have more number of aberrations than those without them. However, the rate of aberrations seen in malnourished children without infections was significantly different from the control aberration frequency.

## DISCUSSION

The results of the present study indicate that the frequency of constitutional chromosomal aberrations was higher in children with severe malnutrition. Further, patients with infections had more number of aberrations than those without infections. This supports the idea that a small amount of chromosome aberrations in malnourished children could have been caused by infections[1]. Since none of the patients in the present study had been exposed to X-rays, the effect of this on the observed frequency can be ruled out. Also, the role of associated defficiency of folic acid, and vitamin $B_{12}$ can be excluded in the present study.

Several factors could have been responsible for the observed chromosomal damage in malnourished children[1]. Possible presence of altered biochemical environment in these children which can produce one or more mutagenic factors, has been suggested[1].

Chromosome aberrations of the kind reported here, have also been seen in a few hereditary cancer-prone syndromes in man, like Fanconi's anemia[17], Bloom's syndrome[4] and ataxia telangiectasia[9]. Incidentally, cells from a few of these syndromes have been shown to have a defect in DNA repair synthesis. It is in this context that a possible DNA repair deficiency in the etiology of chromosomal damage in malnourished children, has been raised[11]. But, the results of our own study on the integrity of DNA repair in the cells from PEM have not revealed any defect[11].

The functional significance of elevated chromosomal damage in PEM is not known. Children with PEM have been shown to have a decreased cellular immunity[16]. The T-cell function has particullarly shown to be defective[7]. Whether increased frequency of chromosomal aberrations reported in PEM could be related to the decreased T-cell function, is not known. Similarly, Bantu children sufferring

from PEM have been shown to have a high incidence of hepatoma[5]. Whether elevated chromosomal damage is related to this, is also not known.

From this study it is also clear that no particular chromosome or chromosome group seems to be more commonly involved in damage in PEM. This observation is in contrast to the data of Armendares et al.[1].

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