SUMMARY AND CONCLUSION

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The present study was concerned with testing the effect of hypercalcaemia on the cell kinetics and chromosomal structure. The blood leukocytes - grown in vitro - were utilized as an experimental model in this respect. The blood samples were obtained from two groups of donors, namely; a male group comprising 8 donors and a female group comprising 7 ones. The leukocytes were separated and the initial cell density was adjusted to fit 200 x 10^3 lymphocytes per millilitre of culture medium utilizing the cetrimide-pronase technique. The lymphocytes were enhanced to undergo proliferation either by the plant lectin phytohaemagglutinin (PHA) or by mixing the bloods of two separate donors (histocompatibility antigenic activation). The calcium ion level was adjusted to fit the ascending tested concentrations through titrating the tested solution against 1/50 N EDTA-buffered solution and the end point was the change of colour from purple to blue.

In experiment I, the leukocyte suspension of each donor was divided into 21 aliquots, one representing a control standard culture and five experimental quadruplets. The calcium ion level was adjusted to 50 µg/ml. in the control culture and to 70, 90, 110, 130 and 150 µg/ml. in the first, second, third, fourth and fifth quadruplets respectively. Four experimental models were employed in each group of quadruplets depending on the time of advent of calcium ion into the culture, namely; simultaneous addition of calcium ion and the mitogen (model A) and five minutes, one hour and six hours after the addition of the mitogen (models B, C and D respectively). In experiment II , five culture series were prepared from 10 donors and the same models were utilized according to the time

lapse between blood mixing and the adjustment of artificial hypercalcaemia.

The index of the proliferative response was estimated by the cell density of the cultures; quantitated at 24, 48, 72 and 96 hours from the inoculation time (Time 0). After 96 hours of incubation, the smears were prepared from each culture tube and were used to estimate the transformation score as well as the chromosomal structure. In chromosomal study, 10 slides were chosen at random - for each culture tube - and 50 metaphases (Experiment I) or 100 metaphases (Experiment II) were examined under the oil-immersion objective, photographed and karyotyped according to the standard human chromosome nomenclature.

The results of the present investigation revealed the following:-

1- In control cultures, the proliferative response was significantly higher in the female donors than in the male ones.

2- Elevation of the calcium ion concentration to 70 µg/ml. (equivalent to 14 mg% in the blood) resulted in an increased response in the male cultures whereas the female proliferative response was suppressed.

3- The suppressive effect of the 90 µg/ml. concentration (equivalent to blood level of 18 mg%) was more evidenced in the female cultures than in those of the male group.

4- Higher calcium ion concentrations (110 and 130 µg/ml.) which are equivalent to blood levels of 22 and 26 mg% were irreversibly toxic to the cells of both groups of donors although the latter concentration was nearly completely lethal to the female cultures.

5- Still higher calcium ion concentration (150 µg/ml.) which is equivalent to a blood level of 30 mg% was completely lethal to both groups of cultures as shortly as 24 hours of incubation.

6- The maximum yield of abnormal karyotypes was recorded with the 90 µg/ml. concentration in both groups of donors in all experimental models. The prevailing anomalies met with in this concentration were tetraploidy and endoreduplication.

7- Chromosomal breaks, however, were highly frequent with higher calcium ion concentrations (110 and 130 µg/ml.).

In conclusion, the data obtained by the present investigation may provide an evidence that the female immunocompetent cells are more susceptible to the drastic effect of high calcium ion concentrations. In addition, chromosomal abnormalities are much more expected in the females when subjected to hypercalcaemia. As an inhibitor of allergic reactions, the ionic calcium is preferably used during the triggering event of the immunocompetent cells; a recommendation which is practically impossible. In addition, the level at which this ion acts meaningfully (18 mg%), is not advised to achieve due to its mutagenic effect on the concerned cells.