isochromatid breaks. Thus, the proportion of marrow cells with breaks in our FA patient (10%) was greater than the average (5%) found in individuals many of whom had diseases known to predispose to chromosome aberrations (Sandberg, 1966). A recent study of bone marrow chromosomes in a male control population found incidences of 3·19% and 0·55% for chromatid and chromosome aberrations respectively and a slightly greater than expected proportion of non-modal (hypodiploid) cells (O'Riordan, Berry, and Tough, 1970). The incidence of chromosome breakage in FA bone marrows may therefore be greater than in normals, but we have not been able to establish this conclusively.

Conclusion

The FA gene is of great interest because of its association with cancer and leukaemia in homozygotes and heterozygotes (Swift, 1971). If the bone marrow chromosomes are representative, it is unlikely that the inherited metabolic defect of Fanconi's anaemia is expressed in vivo primarily through frequent chromosome breaks and rearrangements. The nature of this genetic defect might be elucidated if we could learn what factors are responsible for the increase in chromosome aberrations in cell cultures in vitro from patients with Fanconi's anaemia.

Summary

The chromosomes of bone marrow cells from a patient with Fanconi's anaemia showed a lower aberration frequency than that of either lymphocyte or fibroblast cultures from the same patient. This difference between the aberration frequency in vivo and in vitro may provide a clue to the nature of the inherited metabolic defect underlying Fanconi's anaemia.

We thank Thea Cohen for technical assistance.

SANDRA R. WOLMAN and MICHAEL SWIFT

Departments of Pathology and Medicine, New York University Medical Center, New York, USA

REFERENCES


FANCONI'S ANAEMIA: REPORT OF A PATIENT WITH SIGNIFICANT CHROMOSOMAL ABNORMALITIES IN BONE MARROW CELLS

In Fanconi's anaemia, the presence of frequent chromosomal aberrations in cultured peripheral lymphocytes is now an established phenomenon. In fact, a number of authors (Bloom et al, 1966; Swift and Hirschhorn, 1966; Hirschman et al, 1969), suggest its demonstration as a valuable aid in establishing diagnosis in doubtful cases. On the other hand, very few instances of the above disease have been encountered in which similar changes were seen in bone marrow cells. The following communication describes one such patient.

METHODS

Routine haematological investigations were carried out on blood and bone marrow aspirates according to standard methods.

Chromosome studies were performed on a peripheral leucocyte culture following the method of Moorhead et al (1960). One hundred consecutive metaphases were examined in detail. In addition, a bone marrow specimen obtained by sternal puncture was prepared for karyotyping using a slight modification of the technique of Tjio and Whang (1962). Ninety suitable spreads were photomicrographed and analysed.

CASE REPORT

The patient, an 11-year-old male, was first seen at the paediatric service at the American University Hospital in September, 1966. His symptoms dated back to the age of 4 years, when he started having repeated attacks of easy bruising and bleeding from various sites (nose, urinary tract, rectum, and retina). Meanwhile he was noticed to develop progressive pallor which was not commensurate with the amount of blood loss. There was no history of exposure to drugs or bone marrow toxins.

The family history disclosed consanguinity in the
parents, but was negative for blood dyscrasias or congenital malformations. The patient was born after a full-term, normal pregnancy; his birth weight, however, was subnormal.

On admission the relevant findings were: subnormal weight and stature, a bronze hue of the skin over the face and the trunk, characteristic cafè-au-lait patches over the back, and slightly under-developed genitalia. Mental development appeared to be normal. Initial laboratory data showed a haemoglobin of 3·5 g/100 ml; packed cell volume 11%; white cell count 1900/mm³ with 11% unsegmented neutrophils, 20% segmented neutrophils, 65% lymphocytes, 3% monocytes, and 1% eosinophils; reticulocyte count was 1%, and platelet count 44,000/mm³. A sternal bone marrow aspirate showed decreased cellularity, with the following differential: red cell precursors 36%; myeloid cells 18%; lymphocytes 46%. Few megakaryocytes and platelets were seen. Biochemical data, including blood urea nitrogen, glucose cholesterol, serum proteins, bilirubin, phosphorus, and alkaline phosphatase were all within normal limits. The serum iron level was 238 µg/100 ml and the iron-binding capacity 92 µg/100 ml. Urine and stools examinations were negative. Radiological studies of chest, skull, and long bones and an intravenous pyelogram did not reveal any abnormalities.

The clinical course of the patient over the past 3½ years is outlined in Fig. 1. Initially he was treated with intramuscular testosterone and oral steroids. However, as there was no noticeable haematological improvement, testosterone was replaced by methandrostenolone* with subsequent progressive amelioration of the haemoglobin level. On the other hand, no change was noted in the leucocyte count. Follow-up examination of a bone marrow specimen in April 1970 revealed fair cellularity, with preponderance of the erythroid series but sparsity of megakaryocytes and platelets. Mitosis was seen in approximately 2% of cells, and was most prevalent among red cell precursors (87%). Ten per cent of mitotic figures were abnormal; 3 erythroid cells had anaphase bridges and 4 had chromosome lag, while 3 myeloid cells showed chromosome clumping. A portion of this marrow specimen was used for cytogenetic studies. At present the patient is maintained on the above therapeutic regimen without need for further blood transfusion, although previously 17 units of blood had to be given over a period of 2 years in order to control his anaemia.

---

**TABLE I**

**CHROMOSOME STUDIES**

<table>
<thead>
<tr>
<th>CellsExamined</th>
<th>No. of MitosesAnalysed</th>
<th>% Mitoses with Abnormalities</th>
<th>Type of Chromosome Abnormalities and Number of Mitoses Affected</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Chromatid</td>
<td>Isochromatid</td>
</tr>
<tr>
<td>Peripheral leucocytes, 70-hr culture with PHA* (8.4.1970)</td>
<td>100</td>
<td>20†</td>
<td>22</td>
</tr>
<tr>
<td>Bone marrow direct preparation (1.4.1970)</td>
<td>90</td>
<td>10†</td>
<td>5</td>
</tr>
</tbody>
</table>

* Phytohaemagglutinin—M (Difco). † Excluding gaps. ‡ Probably normal, dividing megakaryoblast.
Chromosome Studies (Table I)

In the peripheral leucocyte culture, chromosome abnormalities were present in 20% of metaphases. The majority consisted of chromatid breaks (11%) while exchanges, rings, and dicentrics were seen in 6 mitoses (Fig. 2). Polyploidy appeared with increased frequency (4%); endoreduplication, on the other hand, was not encountered among any of the cells.

Similar though less frequent changes were found in the direct bone marrow preparation. Of 90 spreads analysed, 9 (10%) had chromosomal aberrations. Here, again, chromatid breaks constituted the main anomalies (Fig. 3), and involved primarily chromosomes of groups B and C respectively. Of particular interest was the presence of a dicentric in one cell and a ring chromosome in another. The significance of these will be discussed later.

In our bone marrow control preparations from 10 subjects with non-haematological disorders, chromosomal rearrangements were not seen in any cell while the range of breaks was 0–4%.

Discussion

There still exists much controversy concerning bone marrow cytogenetics in Fanconi's anaemia.

Swift and Hirschhorn (1966) first reported the presence of chromosomal aberrations, mainly gaps and breaks, in 10% of marrow cells from a female patient with the disease. Likewise, Hirschman and his associates (1969) described changes in 2 brothers with constitutional aplastic anaemia, only one of whom had some of the associated malformations of Fanconi's syndrome. In this patient, 4% of the cells had breaks, fragments or extensive fragmentation, and an additional 4% contained...
dicentrics or rings. In the younger brother, breaks or fragments were seen in 10% of cells. The authors, however, considered the prevalence of breaks in these two patients to be not clearly different from normal. More recently, Guanti, Petrinelli, and Schettini (1971) recorded abnormalities in 20% of bone marrow mitoses from a girl with this disease. On the other hand, such changes could not be demonstrated in a number of other cases investigated (Bloom et al, 1966; Hoenagle et al, 1966; Schmid, 1967; Dosik et al, 1970).

In our patient, a prevalence of 10% is significantly greater than control values encountered here. Furthermore, the presence of dicentric and ring chromosomes in a direct preparation implies that at least some breaks have occurred in vivo. This conclusion is supported by the demonstration of anaphase bridges in smears from the same marrow aspirate.

The fact that our cytogenetic investigations were carried out after 3½ years of combined steroid therapy raises questions as to whether the treatment itself may produce chromosome changes or else modify their prevalence. Evidences from studies in vitro reviewed by Gmyrek et al (1968) and the demonstration of abnormalities in a direct marrow preparation before treatment (Swift and Hirschhorn, 1966) do not favour the first possibility. A modifying effect, however, cannot be ruled out with certainty in the absence of serial bone marrow chromosome studies. One evidence against such an effect on cultured lymphocytes is the persistence of aberration incidence in one of the patients of Hirschman et al (1969), 3 months after reinstitution of the combined treatment.

Whether in-vitro breakage of marrow chromosomes is spontaneous or is induced by extrinsic agents including viruses (Swift and Hirschhorn, 1966), it is conceivable that the genetic deficiencies so produced, provided they are of sufficient magnitude and frequency, may lead to a progressive loss of cells and result in aplastic anaemia. In this respect, the apparent lack of involvement of lymphoid tissues (except the spleen) in spite of a high incidence of breaks in vitro may be attributed, as suggested by Swift and Hirschhorn (1966) to the remarkable length of the intermitotic interval and life span of some small lymphocytes in vitro.

Fig. 3. Chromosome abnormalities in direct bone marrow preparation. Note chromatid gap in a group B chromosome (curved arrow), and acentric fragments (straight arrow).
Summary

In a young male with Fanconi's anaemia, 10% of cells in a direct bone marrow preparation had chromosome abnormalities. Breaks involving primarily groups B and C members constituted the most frequent changes encountered, while 2 cells had either a dicentric or a ring chromosome. Smears from the same aspirate showed anaphase bridges in 3% of mitoses. It is suggested that in this disease chromosome breakage is a process in vivo, and that its occurrence in bone marrow cells possibly contributes to their progressive elimination and ultimate depletion.

MUNIB J. SHAHID, FARID P. KHOURI, and SAMIR K. BALLAS*

Departments of Medicine and Clinical Pathology, American University Hospital, Beirut, Lebanon

REFERENCES


* Presently Research Fellow, Cardeza Foundation for Hematologic Research, Jefferson Medical College, Philadelphia, USA.

Case Report

Down's Syndrome with an Atypical G/G Translocation Derived from Familial Pericentric Inversion in One Chromosome of the G Group

Case Report

The propositus was born on 28 September 1956, the 2nd child of a mother of 30, and a father of 45 years of age (Fig. 1).

The pregnancy and confinement were normal. Birth weight was 4500 g. Psychomotor development was retarded; he sat at 9 months, walked at 18 months, and spoke his first words at 4 years of age. At the first examination, in 1961, Down's syndrome was diagnosed.

In June 1969, when he was 12 years 8 months, he was 131 cm tall and weighed 30·5 kg. Head circumference was 50·5 cm with a cephalic index equal to 0·94; the eyes and the eyebrows were oblique, epicantidial folds were present, and the nasal bridge was flat. There were fissures on the lips and a furrowed tongue. The occiput was typical. The hands were short and the little finger showed clinodactyly. He was tested with the Bühler-Hetzer developmental test, and with the Vineland-

Received 8 May 1972.
Fanconi's anaemia: report of a patient with significant chromosomal abnormalities in bone marrow cells.
M J Shahid, F P Khouri and S K Ballas

doi: 10.1136/jmg.9.4.474

Updated information and services can be found at:
http://jmg.bmj.com/content/9/4/474.citation

**Email alerting service**
Receive free email alerts when new articles cite this article. Sign up in the box at the top right corner of the online article.

**Notes**

To request permissions go to:
http://group.bmj.com/group/rights-licensing/permissions

To order reprints go to:
http://journals.bmj.com/cgi/reprintform

To subscribe to BMJ go to:
http://group.bmj.com/subscribe/