Possible X/Autosomal Translocation in a Girl with Gonadal Dysgenesis

MARIGOLD J. THORBURN, PATRICIA A. MARTIN, and U. N. PATHAK

From the University of the West Indies, Kingston 7, Jamaica

By screening patients with primary amenorrhoea with chromosome studies, many anomalies of the sex chromosomes are detected (Jacobs *et al.*, 1961; Clavero, 1964; Philip, Sele, and Trolle, 1965b; Shearman, 1968; Thorburn and Pathak, 1970). These range from classical Turner's syndrome with 45,XO, mosaicism, 46,XY, apparently normal 46,XX karyotypes, and structural anomalies including deletions, isochromosomes, and fragments. During such a programme over the past 5 years (Thorburn and Pathak, 1970) we found a patient who appears to have a translocation which could be interpreted as either a t(C;C) or a t(X;C). We have been able to find only two similar cases previously reported (Mann *et al.*, 1965; Neuhauser and Back, 1966), and such an abnormality is not mentioned in comprehensive series or reviews of sex chromosome abnormalities (Court Brown *et al.*, 1964; Ferguson-Smith, 1965; Jacobs, 1969). In this paper we present the detailed clinical and cytogenetic findings in this patient and discussion of the possibility of X autosomal translocation. This patient was included in a series of cases of primary amenorrhoea previously reported (Thorburn and Pathak, 1970).

Case Report

A young woman aged 27 came to the Gynaecological Clinic of the University Hospital of the West Indies with a single complaint of failure to menstruate. There had been one episode of scanty vaginal loss at age 18. There was no history of abdominal pain, and intercourse was satisfactory. She was subsequently admitted for investigation.

She was born in a remote country district of Jamaica in October 1941 the 6th child in a family of 8. She was not weighed at birth, and in fact there was no medical supervision of the pregnancy. Her father and mother were 34 and 33, respectively, at the time of her birth, and were unrelated. There was no family history of similar complaints, other congenital or genetic illness, and there

was no history of drug or radiation exposure. Childhood was considered to be normal and she left school at 14.

Examination showed a Jamaican of mixed negro and Caucasian race, with no physical abnormalities except those relating to sexual development. Her height was 171.6 cm., arm span 185.5 cm., and head circumference 57.2 cm. She had minimal breast development, a slightly eunuchoid body habitus (Fig. 1), scanty pubic and axillary hair. The external genitalia were hypoplastic and the vagina long with a small uterus. The adnexae were not palpable. She was of low normal intelligence.

FIG. 1. Patient.

Received 30 January 1970.

Investigations. Routine haematological and biochemical studies were not remarkable. Seventeen ketosteroids and ketogenic steroids were estimated as 4.6 and 5.3 mg./24 hours. A full skeletal survey showed the following abnormalities. The epiphyses of the lower ends of the radius and ulna were not fused, nor were those of the iliac crests (usually fused by age 21). All bones were very porotic. The foramen magnum was asymmetrical and enlarged on the right side. There was congenital fusion of the second and third cervical vertebrae. Intravenous pyelogram showed no abnormality in the urinary tract. A lateral vaginal wall smear showed 5% superficial cells and 95% intermediate cells. A laparotomy was performed on 17 July 1969. This showed an infantile uterus measuring approximately 5 cm. in

length and Fallopian tubes. The ovaries were represented by the typical streak gonads seen in gonadal dysgenesis. No tumours were found. Bilateral salpingo-gonadectomy was performed. She made an uneventful recovery and was subsequently treated with cyclical steroids.

Pathological examination. The portions of tubes measured 5 cm. on each side. In the position of the normal ovary was a fibrous streak overlying rather thick, rich, vascular mesovarial tissue (Fig. 2). Histological examination showed immature Fallopian tubes. The streak area showed fibrous stromal-like tissue with complete absence of oogenesis (Fig. 3). In the mesovarium were blood vessels, primitive tubular structures, and nests of cells resembling interstitial or hilus cells. In some areas these cells seemed to have taken on a cord-like structure (Fig. 4). These appearances were very similar to the streak gonads in other cases of gonadal dysgenesis that we have examined.

Cytogenetic studies. Sex chromatin was examined in 2 buccal smears, using the thionine stain. In 400 cells, 12% were sex chromatin positive. The peripheral blood was processed by a modification of the microtechnique of Arakaki and Sparkes (1963) on 3 occasions: the initial study and for late labelling and autosomal labelling. The culture period was 72 hours, with 1½ hours' exposure to Colcemid (Ciba). Flamedried smears were stained with Giemsa. 25 cells were counted visually and karyotypes were made. Though not detected on initial analysis, karyotypes revealed 3

FIG. 2. Tubes and streak gonads.

FIG. 3. Streak area of gonad showing absence of oogenesis.

FIG. 4. Sub-streak area showing cord-like structures. (H. and E. $\times 320$.)

Fig. 5. Karyotype showing missing members of the C group and 2 translocation chromosomes.

consistent abnormalities (Fig. 5), which were subsequently confirmed by re-examination of this culture, and further examination of unlabelled cells in the autoradiographic studies. These were: (1) 2 medium sized submetacentric chromosomes were missing from the X6-12 or C group; (2) there was an extra 16-sized chromosome; and (3) an extra B-sized chromosome.

This arrangement was interpreted as resulting from a reciprocal translocation between the long arms of 2 autosomes of the C group or an X/autosomal translocation. The rearrangement appeared to be balanced, and it was not possible to detect which members of the C group might be involved.

Examination of the parents' chromosomes showed normal complements in both.

Autoradiographic studies. Autoradiographic studies were carried out on two 72-hour cultures, by the method of Schmid (1963), tritiated thymidine being added at 3½ and 5½ hours before termination of culture. Labelled cells were photographed, the slides degreased by the method of Bianchi, Lima-de Faria, and Jaworska (1964) and restained and rephotographed.

In the first culture (3½ hours), a normal sized X chromosome labelled consistently (Fig. 6). The second culture was not entirely satisfactory. However, in many cells autosomes 4 and 5 could be distinguished from the abnormal translocation chromosome of the same size. The latter showed early replication, especially in the

short arms. The abnormally short translocation chromosome could not be distinguished from the pair number 16.

Discussion

The main interest in this case lies in the interpretation of the chromosomal findings. The clinical management and the question of performing laparotomy with primary amenorrhoea are discussed elsewhere (Thorburn and Pathak, 1970).

Patients with normal stature, infantilism, and dysgenetic gonads without other somatic abnormalities constitute a definite clinical subgroup in cases of primary amenorrhoea. The chromosomal findings can be quite variable including apparently normal complements, 45,X/46,XX mosaicism, 46,XY,46,XXq-, and 46XXpi.

The present case parallels almost exactly that of Mann *et al.* (1965), except that their patients had a t(B;C) and the gonadal status was not known. They presented two possible interpretations of the findings: (a) a *de novo* balanced translocation between 2 autosomes; in their case in the B and C groups, and in our case, 2 members of the C group; and (b) an X-autosomal translocation.

In (a) the translocation would have to be balanced as there was no phenotypic effect other than streak

FIG. 6. Late labelling of a normal sized X chromosome.

gonads. The 2 cases could then be considered examples of primary amenorrhoea with normal sex chromosomes. Autosomal abnormalities are unusual in cases of primary amenorrhoea. In our series of 38 cases we have found one patient with normal ovaries, and atresia of the cervix, who had 46,XX,Dp+, but the significance of this anomaly is in dispute (Court Brown *et al.*, 1966; François, Matton-van Leuven, and Coppieters, 1968). An enlarged No. 1 chromosome was found by Philip, Frydenberg, and Sele (1965a), but other than this anomaly and fragments of unknown origin autosomal abnormalities were not found in a total of 270 patients with primary amenorrhoea in 5 series (Jacobs *et al.*, 1961; Clavero, 1964; Philip *et al.*, 1965; Shearman, 1968; Thorburn and Pathak, 1970), nor are they mentioned in reviews by Ferguson Smith (1965) and Jacobs (1969).

The presence of typical dysgenetic gonads in our case might favour the second hypothesis of an X/autosomal translocation. Though no other definite parallels have been found in man, X-autosomal translocations have been produced in mice (Russell and Bangham, 1959, 1960; Russell, 1961; Ford and Evans, 1964; Lyon *et al.*, 1964). These may behave

in 2 ways: (1) as though part of the autosome had been translocated to the X, and (2) the reverse, where the translocated portion of the X remains active in all cells, as presumably in this case and that of Mann *et al.* (1965). Since we cannot identify which chromosome pairs are involved in the translocation, we cannot speculate on how much material has been lost or gained from each chromosome. The low level of sex chromatin, unlike the case of Mann *et al.*, would also perhaps be in favour of an X being involved. As only a normal X is being inactivated, only half the number of cells will show sex chromatin.

Other cases have been reported in which there is a small possibility of an X-autosomal translocation (Wie Lie, Coenegracht, and Stalder, 1964; Hugh-Jones *et al.*, 1965; Thorburn, Miller, and Dovey, 1967). In these cases, an X chromosome was replaced in some or all cells by a large, late-labelling submetacentric chromosome. However, a complex rearrangement of sex chromosomal material seems more likely. The patient of Neuhäuser and Back (1966) appears to be an intermediate stage between these cases and ours. This infant's karyotype had 45 chromosomes with an extra large dicentric

chromosome and 2 missing C group autosomes. The large chromosome consistently labelled late but only along two-thirds of its length. A bipartite sex chromatin body was observed in 8% out of 20% of positive buccal smear cells. Part of a C group autosome was considered to be lost; this was responsible for the abnormal phenotype. This case suggests that X autosomal translocation may not inactivate the autosome, though the X involved appears to replicate late.

Assuming that an X/autosomal translocation does exist in our case, it is possible that the gonadal dysgenesis is due to loss of a small fragment of the X. However, the presence of streak gonads in patients with normal complements provides a further possibility, that the gonadal dysgenesis is unrelated to the chromosomal abnormality.

Summary

A patient of 27 years with normal stature, primary amenorrhoea, and streak gonads was found to have an abnormal chromosome complement which could be interpreted either as a $t(C;C)$ or a $t(X;C)$. Autoradiography showed consistent late labelling of the normal X but did not aid in identifying the chromosomes involved in the translocation. The possibility of X autosomal translocation is discussed.

We are grateful to our colleagues in chemical pathology and radiology for their assistance, and to Mrs. L. Dawson for the photographic work.

The cytogenetic work was supported by grants from the Standing Advisory Committee for Medical Research in the Caribbean and the Wellcome Trust. Boots Pure Drug Company provided the heparin.

REFERENCES

- Arakaki, D. T., and Sparkes, R. S. (1963). Microtechnique for culturing leukocytes from whole blood. *Cytogenetics*, **2**, 57-60.
- Bianchi, N., Lima-de Faria, A., and Jaworska, H. (1964). A technique for recovering silver grains and gelatin from tritium autoradiographs of human chromosomes. *Hereditas, Genetiskt Arkiv*, **51**, 207-211.
- Clavero, J. A. (1964). Estudio de los cariotipos en las amenorreas esenciales. *Acta Ginecológica*, **15**, 153-159.
- Court Brown, W. M., Buckton, K. E., Jacobs, P. A., Tough, I. M., Kuenssberg, E. V., and Knox, J. D. E. (1966). *Chromosome Studies on Adults*, p. 32. Cambridge University Press, London.
- , Harnden, D. G., Jacobs, P. A., Maclean, N., and Mantle, D. J. (1964). Abnormalities of the sex chromosome complement in man. *Medical Research Council. Special Report Series*, No. 305.
- Ferguson-Smith, M. A. (1965). Karyotype-phenotype correlations in gonadal dysgenesis and their bearing on the pathogenesis of malformations. *Journal of Medical Genetics*, **2**, 142-155.
- Ford, C. E., and Evans, E. P. (1964). A reciprocal translocation in the mouse between the X chromosome and a short autosome. *Cytogenetics*, **3**, 295-305.
- François, J., Matton-van Leuven, M. Th., and Coppieters, R. (1968). Short arm enlargement of a G chromosome. *Acta Geneticae Medicae et Gemellologiae*, **17**, 468-486.
- Hugh-Jones, K., Wallace, S. J., Thornber, J. M., and Atkin, N. B. (1965). Gonadal dysgenesis with unusual abnormalities. *Archives of Disease in Childhood*, **40**, 274-279.
- Jacobs, P. A. (1969). Structural abnormalities of the sex chromosomes. *British Medical Bulletin*, **25**, 94-98.
- , Harnden, D. G., Buckton, K. E., Court Brown, W. M., King, M. J., McBride, J. A., MacGregor, T. N., Maclean, N., Fotheringham, A., and Isdale, M. (1961). Cytogenetic studies in primary amenorrhoea. *Lancet*, **1**, 1183-1189.
- Lyon, M. F., Searle, A. G., Ford, C. E., and Ohno, S. (1964). A mouse translocation suppressing sex-linked variegation. *Cytogenetics*, **3**, 306-323.
- Mann, J. D., Valdiman, A., Capps, S. C., and Puite, R. H. (1965). A case of primary amenorrhoea, with a translocation involving chromosomes of groups B and C. *American Journal of Human Genetics*, **17**, 377-383.
- Neuhäuser, G., and Back, F. (1966). X-Autosom-Translokation bei einem Kind mit multiplen Missbildungen. *Humangenetik*, **3**, 300-311.
- Phillip, J., Frydenberg, O., and Sele, V. (1965a). Enlarged chromosome No. 1 in a patient with primary amenorrhoea. *Cytogenetics*, **4**, 329-339.
- , Sele, V., and Trolle, D. (1965b). Primary amenorrhoea. study of 101 cases. *Fertility and Sterility*, **16**, 795-804.
- Russell, L. B. (1961). Genetics of mammalian sex chromosomes. *Science*, **133**, 1795-1803.
- , and Bangham, J. W. (1959). Variegated type position effects in the mouse. *Genetics*, **44**, 532.
- , and — (1960). Further analysis of variegated type position effects from X-autosome translocations in the mouse. *Genetics*, **45**, 1008-1009.
- Schmid, W. (1963). DNA replication patterns of human chromosomes. *Cytogenetics*, **2**, 175-193.
- Shearman, R. P. (1968). A physiological approach to the differential diagnosis and treatment of primary amenorrhoea. *Journal of Obstetrics and Gynaecology of the British Commonwealth*, **75**, 1101-1107.
- Thorburn, M. J., Miller, C. G., and Dovey, P. (1967). Anomalies of development in a girl with unusual sex chromosomal mosaicism. *Journal of Medical Genetics*, **4**, 283-287.
- , and Pathak, U. N. (1970). Cytogenetic and clinical observations in West Indian patients with primary amenorrhoea. *International Journal of Obstetrics and Gynaecology*, **8**, 43-50.
- Wie Lie, G., Coenegracht, J. M., and Stalder, G. (1964). A very large metacentric chromosome in a woman with symptoms of Turner's syndrome. *Cytogenetics*, **3**, 427-440.