Papers and Originals

Intersexual States and Allied Conditions*

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The first serious attempt to classify human intersexual states was made in 1876 by Klebs, who divided them into true hermaphrodites and pseudohermaphrodites, true hermaphrodites having both testicular and ovarian elements in the gonads, whereas male pseudohermaphrodites had testes but were more or less feminized and female pseudohermaphrodites had ovaries but were virilized.

I. Hormonal Intersexes

Female Pseudohermaphroditism

Almost all the cases of female pseudohermaphroditism are due to congenital adrenal hyperplasia. It is perhaps appropriate to mention that the first clear-cut report of this disorder was made one hundred years ago by the Italian physician de Crecchio (1865). The patient at birth was said by the midwife to be a female and was christened Josephine, but at the age of 4 a surgeon proclaimed the child to be a boy with abdominal testicles, and he was put into male attire and his name altered to Joseph. At the age of 12 he went into service as a valet, and by the age of 18 he had a man's voice, a beard, and a way with the ladies. Eventually at the age of 25 a marriage was arranged, and Joseph was asked to produce his birth certificate. When he discovered he was registered as a female he hesitated to pursue his proposal, and his fiancée jilted him. Thereafter he became a drunkard, a braggart, and a frequenter of cabarets, and when he died at the age of 43 a necropsy revealed adrenal glands as large as kidneys as well as ovaries, normal tubes, and uterus with a vagina that opened into the urethra (Jones and Scott, 1958).

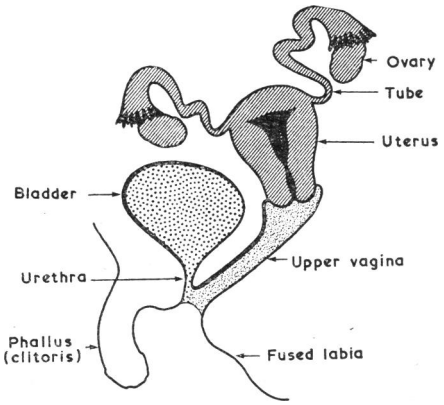

FIG. 1.—Diagrammatic representation of the configuration of the reproductive tract of a case of female pseudohermaphroditism due to congenital adrenal hyperplasia.

The masculinizing influence appears only after complete differentiation of the Mullerian-duct system, so that the tubes, uterus, and upper part of the vagina are normally formed, but the differentiation of the urogenital sinus is incomplete, and that of the external genitalia follows to a greater or less extent a masculine form (Fig. 1). Thus the lower portion of the vagina usually leads into the urethra just before it opens on to the surface, to give the appearance of complete hypospadias, the labio-scrotal folds are fused, resembling an empty scrotum, and the phallus may resemble a penis.

At a stage of embryonic development subsequent to the differentiation of the Müllerian tract there is a variable degree of failure of the enzyme systems that are essential for the biosynthesis of cortisol from progesterone in the adrenal glands. The essential features of this biosynthesis are hydroxylation of the methyl group at C_{21} and the introduction of hydroxyl groups at C_{11} and C_{17} (Fig. 2). The commonest enzyme defect to give rise to the condition is lack of 21-hydroxylase, so that the biosynthetic pathway goes no further than 17α-hydroxyprogesterone, the principal urinary metabolite of which is pregnanetriol. By 1949 Lawson Wilkins had collected 99 cases of congenital adrenal hyperplasia from the literature in contrast to 41 cases of post-natal juvenile adrenogenital syndrome due to tumours of the adrenal cortex. As early as 1937 Butler and Marrian had isolated pregnanetriol from the urine of a woman showing the features of the adrenogenital syndrome, and Bongiovanni *et al.* (1954) were the first to report its increase in the urine of patients suffering from congenital adrenal hyperplasia.

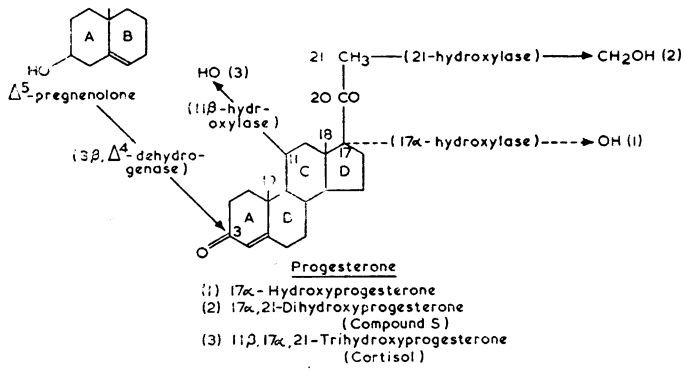

FIG. 2.—The enzymes involved in the biosynthesis of cortisol from progesterone and the conversion of Δ^5 to Δ^4-steroids by 3β-Δ^4-dehydrogenase.

The derivatives of pregnane—namely, the progestogens and the corticosteroids—have the 2-carbon side-chain attached to

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C_{17} as well as the angle methyl groups numbered C_{18} and C_{19}, and are therefore C_{21}-steroids. The derivatives of androstane lack the side-chain, though they possess the angle methyl groups, and are therefore C_{19}-steroids. Lack of 21-hydroxylase, therefore, will not affect synthesis of androgens (C_{19}-steroids) by the adrenal cortex. Furthermore, failure to synthesize cortisol, which is the only adrenocortical hormone that controls the output of corticotrophin from the pituitary, releases enormous quantities of this hormone and accounts for the bilateral hyperplasia of the foetal adrenals, which, however, can produce only androgens, and this they do in great excess. Hence the virilization that is present at birth. Wilkins *et al.* (1950) realized that treatment with cortisone would not only provide the missing glucocorticoids but also curb the release of excessive A.C.T.H. and suppress the adrenal androgen secretion. It would also free gonadotrophins from the pituitary to stimulate ovarian activity and enable feminization of the patient to take place (Fig. 3).

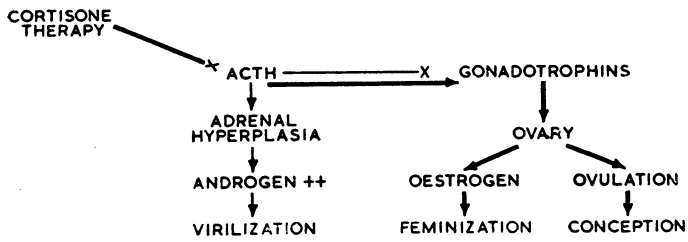

FIG. 3.—Effect of cortisone therapy on a case of female pseudohermaphroditism due to congenital adrenal hyperplasia.

The classical picture is one of early virilization with hirsutism, development of a muscular physique, rapid skeletal growth followed, however, by premature epiphysial closure, leading to stunted adult stature with short arms and legs, and amenorrhoea and lack of mammary development in adolescence. A tendency to salt loss and hypertension characterizes some of the cases. The salt-losing syndrome may complicate severe cases of 21-hydroxylase deficiency or may indicate involvement of the 11β-hydroxylase enzyme system, which, however, is usually accompanied by hypertension, possibly due to accumulation of 11-deoxycorticosterone which fails to undergo 11β-hydroxylation to corticosterone. Visser and Cost (1964) have reported on a congenital salt-losing syndrome due to defective oxidation at C_{18} and consequent failure to synthesize aldosterone.

Finally there is a third enzyme-deficiency block which concerns the conversion of pregnenolone to progesterone, and is due to the lack of 3β-Δ^4-dehydrogenase. Very few cases (about eight) of this so-called congenital lipoid adrenal hyperplasia have so far been described in the literature, though it is difficult to recognize, and many infants have probably succumbed to it undiagnosed. Prader and Anders (1952) gave an account of the condition, and recently a case has been reported by O'Doherty (1964). The condition has so far been invariably fatal, and there is marked salt loss. The adrenals are considerably enlarged and consist almost entirely of cholesterol. There is strong evidence that all cases of congenital adrenal hyperplasia due to enzyme defects are genetically determined, and the likelihood of more than one member of a sibship being affected is sufficiently great to bring to the notice of the parents.

Thus there are three main enzyme defects in the biosynthesis of cortisol that may be responsible for the condition of congenital adrenal hyperplasia presenting as female pseudohermaphroditism. They are:

(1) 21-hydroxylase deficiency, characterized by virilization and possibly the salt-losing syndrome (Bongiovanni *et al.*, 1954).

(2) 11β-hydroxylase deficiency, in which the patients tend to be hypertensive and sometimes pigmented (Eberlein and Bongiovanni, 1956). These two enzyme deficiencies commonly exist in the same patient (Bongiovanni, 1962).

(3) 3β-Δ^4-dehydrogenase deficiency, giving rise to severe adrenal deficiency associated with sodium loss (Bongiovanni, 1961); lipoid adrenal hyperplasia is probably an extreme example in which the adrenal gland is packed with cholesterol that does not even reach the stage of pregnenolone.

Female Pseudohermaphroditism of Non-adrenal Origin

One or two isolated cases have been reported of a woman developing an arrhenoblastoma or other virilizing tumour of the ovary during pregnancy and giving birth to a masculinized female infant—for example, Brentnall (1945)—or having miscarriages in which the foetus showed evidence of female pseudohermaphroditism (Javert and Finn, 1951). The possibility that various sex-hormone preparations therapeutically administered to mothers during pregnancy might masculinize a female foetus has received serious consideration, and by 1960 Wilkins had collected 101 cases of such non-adrenal female pseudohermaphroditism, 35 of the women having received norethisterone, 34 ethisterone, 1 norethynodrel, and 15 a recognized androgen. Two, however, had been treated with pure progesterone, which has not been considered to be androgenic or to give rise to androgenic metabolites. Furthermore, four were treated with stilboestrol, and there were four in Wilkins's series that had had no hormone therapy at all. Incidentally, a case has recently been reported by Lojodice *et al.* (1964) in which 17α-hydroxyprogesterone caproate had been administered to the mother.

Bongiovanni *et al.* (1959), who reported the stilboestrol cases, suggested that this synthetic oestrogen when administered in high doses may give rise to foetal or maternal adrenal hyperplasia, but they pointed out that such occurrences must be very rare, for of 700 diabetic women treated with large doses of stilboestrol throughout pregnancy at the Joslin Clinic not one gave birth to an infant with deformed genitalia. It is even possible that in very rare instances the placenta may produce substances or metabolites with androgenic properties, which would account for the four cases in Wilkins's series that received no hormone therapy, and in other cases reported (Papadatos and Klein, 1954; Wilkins, 1957) in which no apparent masculinizing influence was derived from the mother. Enlargement of the clitoris is an invariable finding, and there is usually partial or complete fusion of the labio-scrotal folds and in some cases persistence of the urogenital sinus with the vagina opening into the posterior urethra: Moncrieff (1958) has described one case in which the vagina was absent. Unlike cases of congenital adrenal hyperplasia, no further masculinization occurs after birth.

Male Pseudohermaphroditism

It is customary to subdivide cases of male pseudohermaphroditism into a masculine, an indifferent, and a feminine type. Though there is no absolute proof, it seems possible that each of these grades is due to a variable degree of failure of the tissues of the body to respond to the androgens, which recent studies indicate are produced in normal amounts by the gonads and possibly the adrenals. The physical characteristics of each individual vary. In the masculine type the defect may be confined to a certain portion of the genital tract, so that incomplete differentiation of the Wolffian-duct system may result in the presence of a rudimentary uterus and tubes, perhaps discovered by chance by a diligent surgeon pursuing an undescended testicle through the internal inguinal ring and disclosing the fimbrial end of a Fallopian tube, or there may be only hypospadias to provide the clue to an enlightened observer.

The indifferent type consists of those cases where the external genitalia present such an anomalous picture at birth that without the aid of nuclear sexing it is impossible to assign

the correct sex to the infant. Many are reared as girls, but when they reach adolescence they first suspect from the failure of breast development, the growth of facial and perhaps body hair, and their muscular physique that they are really men, and some insist, despite the inevitable social embarrassment, on reverting to their true sex. Fig. 4 shows the usual disposition of the external genitalia with the penis anchored by chordee and crowned by a bifid scrotum, each testis lying on either side in the labio-scrotal fold. The object of the plastic surgeon is to construct a scrotum below the freed penis and bring the testes into their correct position.

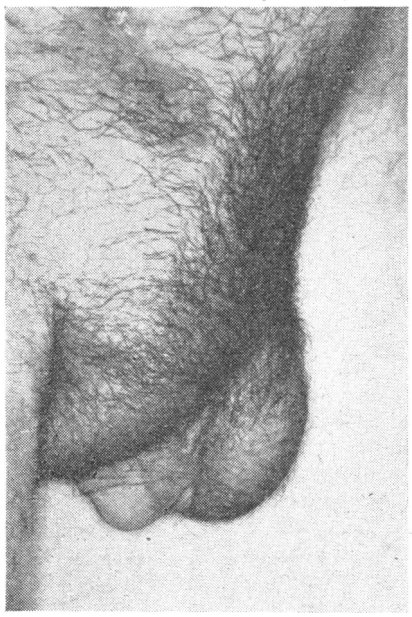

FIG. 4.—Disposition of the external genitalia in a case of male pseudohermaphroditism.

Testicular Feminization

Morris (1953), who suggested the term "testicular feminization" for the feminine type of male pseudohermaphroditism, laid down the following criteria for the syndrome: (1) female habitus with a tendency to eunuchoidism ; (2) normal or somewhat large female breasts with poorly developed nipples ; (3) absent or scanty pubic and axillary hair ; (4) normal female external genitalia with a normal clitoris and a blind vagina (Fig. 5) ; (5) absent or rudimentary internal genitalia ; (6) gonads which histologically resemble undescended testicles and produce androgens and oestrogens, and which may be intra-abdominal or lie anywhere along the course of normal testicular descent, and may therefore be found in the inguinal canals or even in the labia—these testes have a high risk of becoming malignant, and Morris and Mahesh (1963) say it may rise as high as 22% in patients over the age of 30 ; (7) there is a strong hereditary tendency ; and (8) the nuclear sex is chromatin-negative and the karyotype shows a modal figure of 46 chromosomes with a normal male XY sex chromosome pattern.

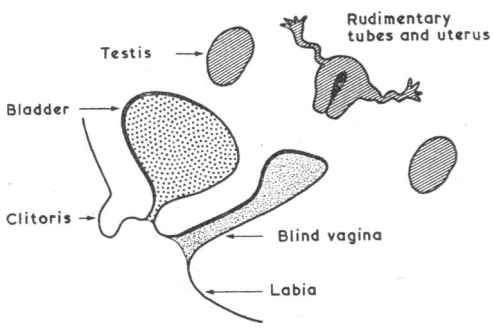

FIG. 5.—Diagrammatic representation of the configuration of the reproductive tract of a case of testicular feminization.

Attention was originally called to this syndrome by Goldberg and Maxwell (1948), who by then had collected eight cases from the literature, so that at one time it was referred to as the Goldberg-Maxwell syndrome (Bishop, 1954). By 1953 Morris had collected 82 cases, including two of his own, and had pointed out the great tendency for the condition to prevail not only horizontally, among sibs, but also vertically in a family tree.

Carrier females may show slight traits of the condition, such as rather scanty sexual hair or a delayed menarche (Philip and Sele, 1965). The condition is strongly inherited in a manner that suggests either a sex-linked recessive or a sex-limited dominant mode of transmission through unaffected females (Jagiello and Atwell, 1962). Neher *et al.* (1965) state that nearly 190 cases of the syndrome have been reported up to date. It is widely held that, in the male during embryonic development, a male evocator in the early stages and the action of testosterone secreted by the foetal testes in later stages are essential for the suppression of the Müllerian-duct elements and the differentiation of the Wolffian-duct derivatives and the external genitalia. The presence in the foetal testicles of a factor that inhibits Müllerian-duct differentiation would account for the absence of tubes and uterus, and failure of the androgenic properties of the testosterone of the foetal testis would account for the feminization of the urogenital sinus, resulting in a blind vagina, and of the external genitalia. At adolescence the non-action of the testicular androgens and the normal functional activity of their oestrogens would account for the development of female secondary sex characters.

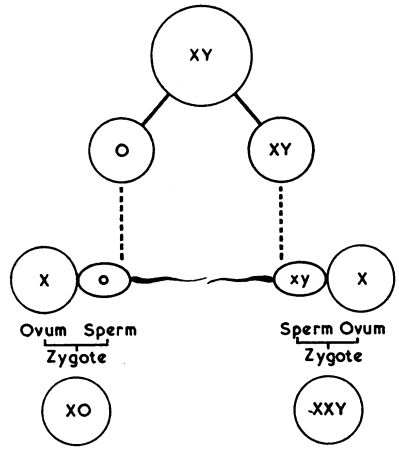

FIG. 6.—Meiotic non-disjunction during spermatogenesis (first division).

It has been reported (Southren *et al.*, 1965) that the plasma testosterone concentrations of three cases of the syndrome were within the normal range for adult men, and removal of the gonads caused a sharp decrease to a level somewhat above the normal female and castrate male range, which suggests that the adrenal cortex contributes in part to the circulating level of testosterone. The *in vitro* studies showed that the gonadal tissue was capable of synthesizing testosterone from progesterone and androstenedione, and oestrogens from testosterone and androstenedione. Their observations support the suggestion that the syndrome is due to insensitivity of the tissues to circulating androgen. There are now several reports—referred to, for instance, by Hauser (1963)—of studies of urinary oestrogen levels before and after castration which make it clear that the oestrogen is synthesized only in the testicles. Removal of the gonads is often followed immediately by hot flushes, and the current consensus of opinion is that the gonads should be

left *in situ* until female secondary sex characters have developed satisfactorily and then be removed because of the risk of malignant changes.

Thus the present concept of male pseudohermaphroditism is that it is due to a relative or complete insensitivity of the tissues to endogenous, and indeed exogenous, androgen ; for high doses of exogenously administered androgens applied systemically or locally will not induce sexual hair growth in cases of the complete syndrome. In incomplete cases only relative failure of androgenization of the genital tract is evident, so that the spectrum of this "non-virilizing testis syndrome" ranges from the classical picture of testicular feminization through the indifferent to the masculine type of male pseudohermaphroditism. There is at the moment little, if any, evidence of any abnormality of sex-chromosome pattern, and all the cases are thought to have a normal male XY-46 chromosome pattern.

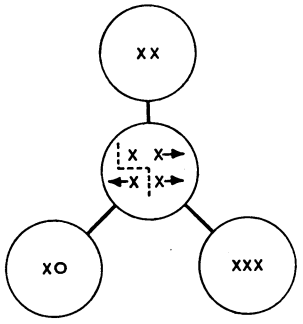

FIG. 7.—Mitotic non-disjunction at the first cleavage division of the female zygote, giving rise to an XO/XXX mosaic.

Thus female pseudohermaphrodites are virilized by androgenic influence developing during foetal life, and male pseudohermaphrodites are feminized by defective response to normal levels of endogenous androgen.

II. Sex Chromosomes

The nucleus of every cell except certain germ cells contains the diploid number of 22 pairs of autosomes and one pair of sex chromosomes, XY in males and XX in females, making a total of 46. Each cell reproduces itself by mitotic division into two daughter cells which exactly resemble the parent cell and contain 46 chromosomes. In the course of gametogenesis meiotic division takes place, one chromosome of each pair passing into each of the two daughter cells and giving rise to the haploid number of 22 autosomes and one sex chromosome, which in the case of the female is always X but in the male may be X or Y. Fertilization of the ovum restores the diploid number of 46, the sex-chromosome pattern being XY or XX, depending on whether the ovum was fertilized by a Y-bearing or an X-bearing sperm.

In 1949 Barr and Bertram observed a deeply staining mass in the nuclei of the neurones of the hypoglossal nerve of female cats ; this mass was absent in the nuclei of male cats. This arrangement was found to exist in several species, including the human. The mass proved to be an X chromosome tightly coiled and thereby taking up stain excellently—heterochromatic. It eventually turned out that in every nucleus there is always one, but only one, X chromosome that carries out the genetic duties of producing and releasing R.N.A., and that in the process its D.N.A. is stretched out and extended and therefore does not take up stain—it is euchromatic. Thus the chromatin mass represents an inactive X chromosome, and consequently there is always one less chromatin mass than the number of X chromosomes. So females are chromatin-positive and males are chromatin-negative.

Tjio and Levan (1956) and Ford and Hamerton (1956) devised a technique by which the individual chromosomes in a nucleus in metaphase can be separated from one another. The chromosomes are then microscopically magnified and photographed, and the image of each chromosome is cut out and pasted on a sheet, the chromosomes being arranged in pairs according to the length of their arms and the position of their centromeres. This is known as a karyotype, and from its study one is able to identify directly or by inference the number of X and Y chromosomes.

This technique enabled cytogeneticists to study sex-chromosome anomalies. In the first place it was found that the modal number of chromosomes, in karyotyping a suitable sample of metaphase nuclei, might be more, or one less—namely, 45—than the normal diploid number of 46. This aneuploidy indicates a sex-chromosomal anomaly. Such anomalies are caused by one of the following mishaps that may take place during cell division: non-disjunction, X-chromosome deletion, isochromosome formation for the long or short arms of the X chromosome, or mosaicism due to non-disjunction or to anaphase lagging.

Non-disjunction may occur during meiotic or mitotic division. In its simplest form the two sex chromosomes fail to separate, and consequently both appear in one of the daughter cells and no sex chromosome is found in the other daughter cell. Should this occur during spermatogenesis and the X ovum be fertilized by an XY-bearing sperm the sex-chromosome pattern of the zygote will be XXY—such as commonly occurs in Klinefelter's syndrome. Should a sperm carrying no sex chromosome fertilize an X ovum the zygote will be XO, which is the pattern of the classical chromatin-negative Turner's syndrome with a modal chromosome count of 45 (Fig. 6). In mitotic division replication of the chromosomes occurs before the cells actually divide, so that in a female nucleus just before division there will be two pairs of Xs. If non-disjunction should take place in the first or second cleavage division of the zygote three X chromosomes could pass into one of the daughter cells and only one into the other (Fig. 7). During gametogenesis there are actually two meiotic divisions, and non-disjunction could occur in both of them. The fact that the second meiotic division behaves in a similar fashion to mitotic

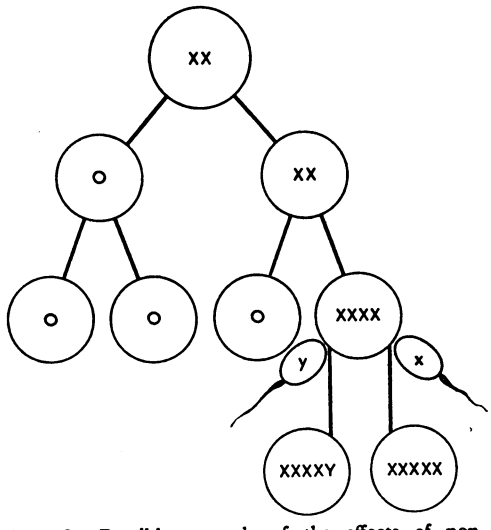

FIG. 8.—Possible example of the effects of non-disjunction at the first and second meiotic divisions.

replication results eventually in four daughter cells, each with one or other of the original parent sex chromosomes. Multiple Xs or Ys could result from non-disjunction occurring in both meiotic divisions (Fig. 8).

During the actual process of nuclear division there may be loss or destruction of one of the arms of a chromosome. This may occur to an X chromosome, and if the normal pattern

is female the daughter cell will have an X deleted-X(X$_{\Delta}$) karyotype. This would be expected to diminish the overall XX influence on the tissues.

If the centromere splits horizontally instead of vertically both the long arms will enter one daughter cell and the short arms the other. The centromeres of the resulting chromosomes will be metacentric and the arms will be of equal length. This is known as an isochrome, respectively of the long or the short arm of the X (for it is usually an X chromosome that is affected) (Fig. 9).

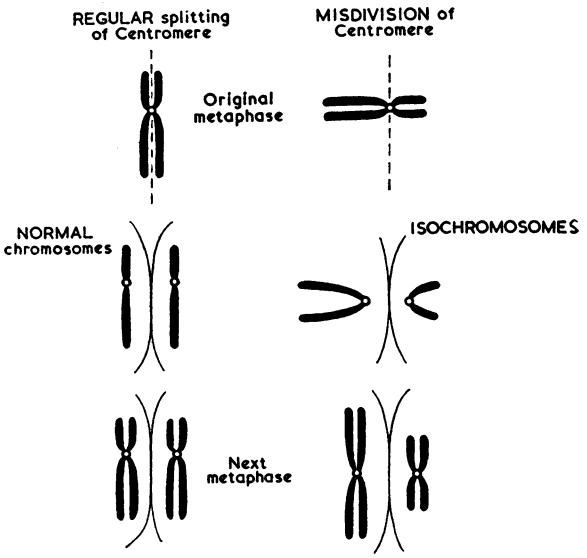

FIG. 9.—Origin of an isochromosome through misdivision of the centromere during mitotic division.

Errors of chromosomal division may occur during the first, or an early, cleavage (mitotic) division of the zygote. Since in mitotic division all the successive generations of the parent cell exactly resemble that cell so far as its sex-chromosome pattern is concerned, non-disjunction in the early cleavage divisions may give rise to two or more cell lines (Fig. 10), each rapidly multiplying as embryonic development proceeds, so that eventually metaphase chromosomal studies of post-natal tissues, such as leucocytes in blood cultures, fibroblasts in skin biopsy specimens, bone-marrow cells, and, most significant of all, cells undergoing gametogenesis obtained by testicular and sometimes ovarian biopsy may disclose the existence of more than one cell-line, which are referred to as mosaics.

Though mosaicism is usually due to post-zygotic non-disjunction, it may result from anaphase lagging. This means that in the penultimate or anaphase stage of mitotic division, when the chromosomes of each pair have migrated to the opposite poles of the cell, one sex chromosome is functionally unprepared for the migration and lags behind and eventually is not included in the final or telophase of the mitotic division. Anaphase lagging may also, therefore, give rise to mosaics, such as XX/XO (Fig. 11).

III. Chromosal Intersexes

The two main groups of chromosomal intersexes are those due to dysgenesis of the ovaries and those due to dysgenesis of the testis.

Turner's Syndrome

The classical case of the former is Turner's syndrome with an XO/45 modal chromosome number and three principal characteristics—sexual infantilism, short stature, and congenital anomalies (Fig. 12).

Sexual infantilism is manifested by primary amenorrhoea; lack of breast development with widely spaced nipples and grossly hypoplastic areolae; scanty sexual hair, which grows fairly profusely on oestrogen therapy; infantile external genitalia, uterus, and tubes; and "streak" gonads lying in the broad ligaments and containing no ovarian follicles but consisting simply of fibrous tissue. Urinary gonadotrophin levels are usually raised.

Short stature is invariable and the patients never attain a height of more than 5 ft. (152 cm.). The birth weight tends to be low, and Acheson and Zampa (1961), examining our cases at Guy's, found that up to the age of 10 the bone age was only slightly retarded, but that skeletal maturation ceased at the time of expected puberty and epiphysial closure failed to occur, except under subsequent treatment with oestrogen.

Congenital anomalies vary considerably in their incidence and nature. Webbing of the neck must be present to justify the terminology of Turner's syndrome (Turner, 1938), though it is not confined to this condition, and in a study of 75 cases

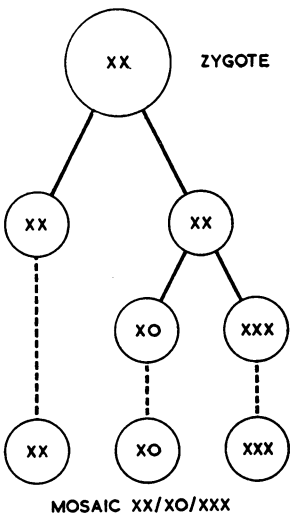

FIG. 10.—Mitotic non-disjunction of the second or subsequent cleavage divisions of the zygote, giving rise to a triple mosaic.

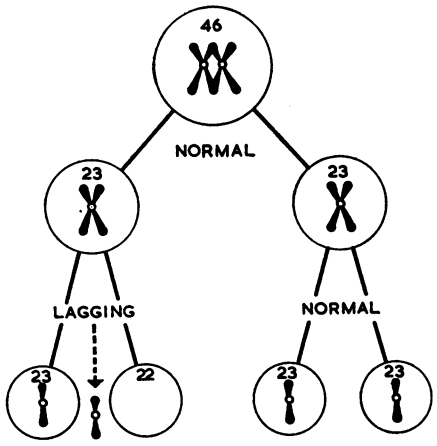

FIG. 11.—Anaphase lagging giving rise to an XO/XX mosaic (second meiotic division).

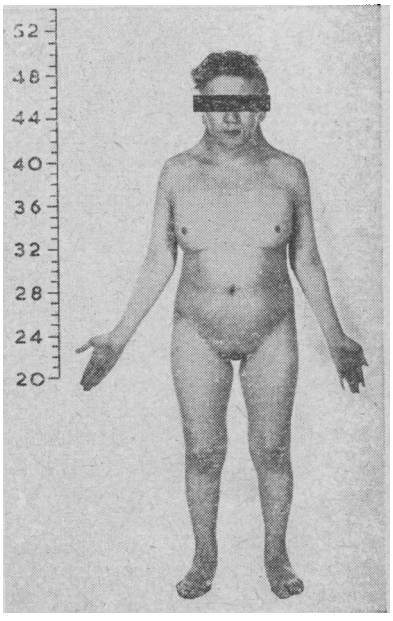

FIG. 12.—Turner's syndrome.

of Turner's syndrome and allied conditions reported on by us in 1960 (Bishop, Lessof, and Polani) we recognized the webbing syndrome of Bonnevie-Ullrich (Ullrich, 1930), occurring both in females and in males, and the Klippel-Feil syndrome of synostosis of the cervical vertebrae sometimes associated with webbing (Klippel and Feil, 1912).

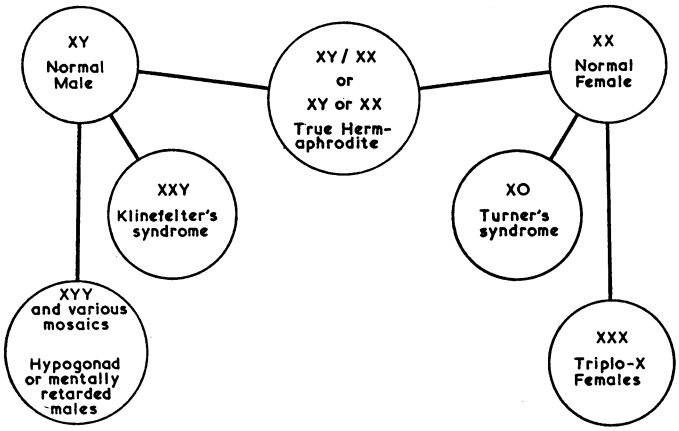

FIG. 13.—Sex-chromosome patterns in cases of intersex and allied disorders.

The facies is characterized by flattened nose-bridge, exaggerated epicanthic folds, antimongolian eye-slant, sometimes with ptosis, depressed corners of the mouth, and low-set ears with auricular malformations. An excessive number of pigmented moles is a prominent feature, and in a quarter of all the patients there is congenital lymphoedema of the legs, feet, and hands, which disappears in a few months or years after birth.

Congenital cardiovascular anomalies are common, and especially coarctation of the aorta, which in the general population is much more frequent in males than females. In our series of cases coarctation occurred in one-third of the classical examples of chromatin-negative Turner's syndrome, and this was the only congenital vascular anomaly we found in these cases.

Renal anomalies, especially horseshoe kidney, which in the general population is commoner in males than females, were fairly often found, and horseshoe kidney or double ureters were found in 25% of our cases of chromatin-negative Turner's syndrome.

The majority of cases of chromatin-negative ovarian dysgenesis with or without webbing have 45 chromosomes and a single X sex chromosome (XO pattern), but there is a much smaller subgroup of mosaics with one of the following patterns: XO/XX, XO/XY, or XO/XYY. It will be seen that the XO pattern is common to all these cases, and is almost certainly the cause of the streak ovaries. The chromatin-negative XO cases with webbing constitute classical Turner's syndrome. If webbing is absent there tend to be fewer and less typical congenital anomalies.

Chromatin-positive Ovarian Dysgenesis

Chromatin-positive cases of ovarian dysgenesis fall into two main groups: mosaics with normal chromosomes, and cases in which one X chromosome is abnormal in shape, being either an isochromosome or a deleted X. In each of these groups there is present a cell line in which the full XX influence needed for normal ovarian development is lacking, and hence the ovarian dysgenesis. The chief mosaic patterns found have been XO/XX, XO/XXX, or XO/XX/XXX. The chromatin-positive cases tend to have relatively few congenital anomalies, and may be difficult to diagnose clinically. Some even have incomplete dysgenesis, and there may be a few ovarian follicles associated with the occurrence of an occasional menstrual period during adolescence. Cases have been described, originally by Gordan *et al.* (1955), of ovarian dysgenesis with androgenic manifestations such as phallic enlargement, due to some testicular elements in the streak. For some years now Polani and his team and I and my assistants have been studying the chromosomal pattern of patients with primary and secondary amenorrhoea, and this will form the subject of a future publication. At one stage we had collected 79 cases of ovarian dysgenesis, of which 32 were chromatin-positive. Seven of these had a normal XX chromosome pattern, five were XO/XX, and five were XX. The rest were mosaics containing XO or complexes containing an abnormal X. It is difficult to explain the seven normal XX cases, though of course one can never be absolutely certain they were not undisclosed mosaics, or had merely hypoplastic or even normal instead of streak ovaries, if these had not been submitted to actual visual examination, as we have found in some of our apparent cases of pure gonadal dysgenesis.

Male Turner's Syndrome

Male Turner's syndrome, associated with inadequate testicular development, short stature in some cases, webbing of the neck, and other congenital anomalies, has been reported by numerous authors—for example, Sharpey-Schafer (1941). All the cases, except one (Schoen, 1965), reported in the literature that have undergone chromosome studies have been mosaics with XO/XY or XO/XY/XYY. The exception was a normal 46/XY, but it could have been a mosaic. The presence of an XY pattern was probably responsible for the presence of testes, though the XO influence was probably responsible for the defective development of the testes. They usually showed complete germinal cell aplasia.

Pure Gonadal Dysgenesis

We have been especially interested in a variant of ovarian dysgenesis in which the failure of ovarian development with the accompanying lack of secondary sexual development is the only manifestation of the syndrome. The stature is not short —in fact, these women may actually be tall—and there are no congenital anomalies. This is therefore known as "pure" gonadal dysgenesis. Brøgger and Strand (1965) have collected 38 cases from the literature.

Two hundred and four cases of ovarian dysgenesis and allied disorders have been or are in process of being studied in Polani's cytogenetic unit at Guy's. Of these 204 cases 18 conformed to the criteria we had laid down for a diagnosis of pure gonadal dysgenesis—namely, primary amenorrhoea, generally insufficient development of secondary sex characters well after the age of puberty, a raised level of urinary gonadotrophins, and radiological evidence of relatively mild retardation of bony development. The height was 5 ft. (152 cm.) or more with either normal or eunuchoid body proportions. We realize that the diagnosis can be finally confirmed or refuted only if the gonads can be visualized either by culdoscopy or laparotomy, and we feel that this procedure is justified—indeed is in the interest of the patient—for if she is chromatin-negative, exploration is essential to exclude a diagnosis of testicular feminization and therefore removal of the gonads on account of the high risk of malignancy. If the patient is chromatin-positive, delayed puberty could not otherwise be excluded, and it would not be possible to give prognostic advice.

Of the 17 possible cases of pure gonadal dysgenesis nine have had a laparotomy and three a culdoscopy. Of these 12 five presented with gonadal streaks, thus confirming the diagnosis; the sex-chromosome pattern was XY in two cases and

XX in three cases. In two other cases there was a streak on one or both sides and a cystic tumour not necessarily associated with the ovary on the other, and the chromosomal pattern was XO/XY. The remaining five presented with slightly hypoplastic or normal ovaries and sex-chromosome patterns of XX, and were therefore not cases of pure gonadal dysgenesis. Thus of the 17 cases under consideration seven were proved to be cases of pure gonadal dysgenesis, and the sex-chromosome pattern was XX in three of them.

Polani (personal communication, 1965) has now collected from the literature—including 79 cases of ours, not all published—282 cases of ovarian dysgenesis. These consisted of series of a reasonable number of cases, and did not include reports on single patients. Among these 282 cases there were 23 examples of pure gonadal dysgenesis; 7 were chromatin-negative, 6 being XY and 1 XO/XY; 16 were chromatin-positive, and 9 of these were XX, 1 XA, 3 XO/XX, 2 XO/XA, and 1 XO/XX/XXX. He has carefully examined the ovarian streaks microscopically in a number of our cases of gonadal dysgenesis, and those from chromatin-negative XO cases consisted simply of non-functional fibrous or connective tissue, whereas streaks from other cases showed some epithelial cells. Perhaps it is just a matter of degree, the XX cases lacking some gene that plays a part in the development and differentiation of a normal ovary. The XY cases are also interesting, and Kinch *et al.* (1965) suggest that these are really cases of gonadal agenesis which are, however, genetically male. Jost (1947) showed that if one extirpated the gonads of a rabbit foetus at an early stage of embryonic life before gonadal differentiation had begun the rabbit was eventually born with a female phenotype, even though it may have been destined genetically to be a male. This is because it lacks the male evocator and testosterone secreted by the foetal testis. There is no corresponding female evocator, nor does the foetal ovary produce oestrogen, so that if the gonads are ovaries, or are absent, the phenotype is female. These XY cases of gonadal dysgenesis (or agenesis) might therefore be described as Jost's syndrome.

Multiple X Syndromes in Women

Geneticists familiar with the chromosal variations encountered in the fly, *Drosophila melanogaster,* used to refer to the XXX variety as "super-female" (Morgan *et al.,* 1925). There was therefore slight disappointment when the first human case to be reported, by Jacobs *et al.* (1959), was that of a rather undistinguished but otherwise physically normal woman with secondary amenorrhoea. This condition (now referred to as triplo-X), which is rather common (about 1 in every 1,000 female births), and its rarer variant, tetra-X, be detected only by means of routine buccal smear examinations revealing nuclei with two or three chromatin masses in association with a normal female phenotype, with perhaps slightly underdeveloped secondary sex characters and secondary amenorrhoea developing at an early age, but not necessarily infertility. Some of these women have borne children with normal nuclear sex, whereas one might have expected secondary non-disjunction. The most interesting feature of these multiple X individuals, both female and male, is a tendency for them to be educationally subnormal or even mentally defective.

Klinefelter's Syndrome

The main clinical manifestation of a chromosomal anomaly in males is Klinefelter's syndrome (Klinefelter *et al.,* 1942). The boy grows and develops normally until puberty. Then one or more of the three classical features may become apparent: the testicles remain small and are usually firm in consistency, gynaecomastia may develop, and there may be signs of eunuchoidism such as long arms and legs and poor sexual hair and beard growth. The testes are small because there are few seminiferous tubules, containing only Sertoli cells, or even completely hyalinized, though there is an excessive amount of interstitial tissue. The patients are frequently detected at a fertility clinic, where they are found to have aspermia and small testes, and the consequent examination of a buccal smear indicates that they are chromatin-positive and that their sex-chromosome pattern is XXY. Chromatin-negative (XY) cases have been described, but in many of these cases testicular biopsy reveals peritubular fibrosis, possibly resulting from previous inflammatory disease, so that the condition is not genetically determined. Other variants are XXXY (triplo-X,Y), XXYY (double X, double Y), and XXXXY (tetra-X,Y). In addition, a number of mosaics have been identified. Ford *et al.* (1959) reported the first human mosaic, which was a case of Klinefelter's syndrome with an XXY/XX sex-chromosome pattern. Other mosaics are XXY/XY, triplo-X,Y, and tetra-X,Y/triplo-X,Y. The patient with classical Klinefelter's syndrome is usually not educationally subnormal or mentally defective. The XY/XXY type may be mentally defective, and patients with more than two Xs are very likely to be.

XXXXY Syndrome

The tetra-X,Y condition, of which eight cases have been reported, presents a rather special syndrome. Mental retardation has occurred in every case. The facies is sufficiently typical to be easily recognizable, showing epicanthic folds, widely set eyes, and slanting of the palpebral fissures (Joseph *et al.,* 1964). The penis is small, the testes are impalpable, and the scrotum is rudimentary. Skeletal anomalies consist of kyphosis, lordosis, and radio-ulnar synostosis—also reported in a triplo-X,Y male (Ferguson-Smith *et al.,* 1960). In most of the cases the little finger curves in. The case of Joseph *et al.* had a patent ductus arteriosus, and so did the case of Fraccaro *et al.* (1960).

XO/XY Syndromes

The XO/XY mosaic seems to be a meeting-point in the various varieties of chromosomal intersex. It has been reported in cases of true hermaphroditism, in a female patient with intra-abdominal dysgenetic testes, and in one of our cases of pure gonadal dysgenesis, as well as in males with intra-abdominal testes lying in the position of the genital ridges and in one case of male pseudohermaphroditism. It would seem that either the XO component gives rise to a female manquée or the XY component is diluted by the X of the XO component and is responsible for the undescended and dysgenetic testes of a phenotypic male. Indeed, the combination may actually give rise to a mixture of ovarian and testicular elements, as in the cases of true hermaphroditism.

True Hermaphroditism

In cases of true hermaphroditism one gonad may be a testis and the other an ovary. A case has even been reported in which a testis and an ovary were present on both sides of the body (Clayton *et al.,* 1957). Or else there is a testis or an ovary on one side and an ovotestis on the other, or bilateral ovotestes. There is a local tendency for the phenotypical characteristics to follow the genotype, and one case has been described with an ovary on one side with marked female phenotypical characters such as breast development and labia, and a testis on the other side with increased muscularity, a male nipple and areola, and a scrotal fold (Brachetto-Brian *et al.,* 1943). Blank *et al.* (1964) reported on the 16 cases in the literature up to date that had been submitted to chromosomal studies. Twelve had a normal female 46/XX pattern, and only one had a normal male 46/XY karyotype. One was an XX/

triplo-X mosaic, another an XX/XY mosaic, and another was a triple mosaic XX/XXY/XXYY. In addition, there were two unproved cases in so far as the gonads had not been examined, and they had an XY/XO pattern. Their own case, the seventeenth, was an XX/XXYY mosaic. Here again is the curious fact, previously encountered in chromatin-positive ovarian dysgenesis and pure gonadal dysgenesis, that an appreciable number of cases have an apparently normal female sex-chromosome pattern, XX.

XX/XY Chimeras

The XX/XY combination raises the question of chimeras and the possibility of double fertilization of an ovum by an X-bearing sperm and a Y-bearing sperm. Bain and Scott (1965) reported a singleton female somewhat resembling a case of testicular feminization but having gonads consisting of a streak on one side and a disgerminoma on the other. The sex-chromosome pattern was XX/XY, and the authors refer to the case as one of mixed gonadal dysgenesis. Of the nine previous cases two were opposite-sex twins with no intersexual traits but with mixed red-cell populations suggesting an interchange of haemopoietic tissues between the two foetuses *in utero* ; in other words, twin chimeras. Six cases, including their own, three of which were true hermaphrodites, one a male with gynaecomastia and another a male with hypospadias, were considered to be due to dispermic conceptions, and the remaining two cases, a chromatin-negative case of gonadal dysgenesis and a chromatin-negative true hermaphrodite, were thought to be due to mitotic errors in an XY zygote.

Conclusion

It appears that female pseudohermaphroditism is due to the influence of androgens on the foetus, and male pseudohermaphroditism to resistance of the tissues to normally circulating androgens. The intersex states produced by sex chromosomal errors indicate in general that the presence of a Y chromosome is necessary for a testis to develop, though if the Y chromosome has to compete against more than one X chromosome the testis becomes dysgenetic and usually contains no germinal epithelium (Fig. 13). Further multiple Xs are usually associated with mental deficiency. Lack or abnormality of the second X chromosome gives rise to ovarian dysgenesis, gonadal aplasia leads to development of a female phenotype, even though an XY chromosome indicates that the individual was destined to be a male, and XX/XY patterns are sometimes due to fertilization of a binucleate ovum with an X-bearing and a Y-bearing sperm. But there is no really satisfactory explanation of cases of true hermaphroditism, ovarian dysgenesis, and pure gonadal dysgenesis with an apparently normal XX chromosome pattern.

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