LINKAGE AND CROSSING-OVER IN THE HUMAN SEX CHROMOSOMES

By THEODORE WHITE, PH.D.

(Fellow of the University of Wales)

(Statistical Appendix by Prof. J. B. S. HALDANE, F.R.S.)

(With Eleven Text-figures)

THE outstanding feature of modern research in human genetics is the emphasis laid by many workers on the necessity of obtaining data concerning the "linkage" of genes, evidence of which was first obtained for human beings by Madlener (1928). The results of the work by Morgan and his collaborators on *Drosophila melanogaster* are sufficient evidence of the importance of this phenomenon in relation to a proper understanding of the mechanism of heredity.

Haldane (1936), by statistical analysis of the data accumulated by previous workers regarding a group of hereditary defects, came to the conclusion that the data required the assumption that crossing-over of the X- and Y-chromosomes of man was an important factor in human heredity. Statistically, as was indicated by Fisher (1936), this conclusion can be regarded as a valid interpretation of the facts but, as Haldane (1936) himself indicated, no clear-cut case of chromosome crossing-over was then available in respect to human heredity. In the case of *Drosophila*, the study of abnormal ratios in the inheritance of linked defects has led to the construction of extremely detailed maps of the chromosomes of this organism and Haldane (1936), as a result of the work noted above, succeeded in mapping provisionally the loci of six genes on the pairing segment of the X-chromosome in man. He pointed out also that gene-linkage in man would undoubtedly be most easily recognized in the case of genes carried on the sex chromosomes and suggested, in view of the relatively high incidence, of colour-blindness in the general population, that all persons known to any investigator to be suffering from sex-linked hereditary defects should be examined for colour-blindness. Such an example of colour-blindness in association with another sex-linked defect is available in the present work.

The study of the hereditary transmission of ocular defects in man has already led to the discovery of anomalies of which not the least important
is the fact that many of these defects are characterized by the; existence of two distinct modes of genetic transmission. Punnett (1933) indicated that this situation has no close parallel in animal genetics, for the defects may be transmitted as simple Mendelian dominants or as sex-linked recessives, without any clinical differentiation between the types from these two sources. The existence of this anomaly is well instanced in the studies of night-blindness which are summarized up to 1932 by Bell (1932). The defects classed under this title fall into three major categories, the first including that form of night-blindness which results from vitamin A deficiency. It is capable of clinical treatment in that the defect disappears on restoring a normal vitamin balance in the diet, and is therefore genetically of little interest. The second category concerns the defect Retinitis pigmentosa, identified readily in that it is progressive, generally resulting in total blindness, and is characterized by distinct fundus changes which are readily observed by ophthalmoscopic examination of the affected person.

The third category, which is important in relation to the present work, concerns congenital stationary night-blindness. This form of the defect may be distinguished from that in the two preceding categories in that it is present from birth, is non-progressive, is not cleared up by vitamin A treatment, and in that ophthalmoscopic examination reveals no fundus changes that can be regarded as typical of the defect. This category is further subdivided by Bell (1932) according as to whether or not the defect is transmitted as

(i) a Mendelian dominant, appearing in both males and females,
(ii) a sex-linked recessive, appearing in the males only but transmitted by the females.

This latter subcategory is further divided according as to whether or not

(a) the defect appears alone,
(b) the defect is accompanied by myopia.

Myopia itself and colour-blindness, with which two defects the present work is also concerned, are again known to be transmitted in sex-linked fashion (cf. Bell, 1932), although this is not universally true of myopia.

Bell (1932, p. 195), in discussing these defects, stated: "When we look for other defects or anomalies characteristically associated with colour-blindness we can find no evidence at all that such exist... Associated eye defects appear to be extremely rare and of no apparent significance... our information is incomplete and may be misleading..."
nevertheless, other eye defects have been too frequently sought for to be
overlooked if at all frequent and we are inclined to the view that colour-
blindness is probably not characteristically associated with other heredi-
tary defects. We might even tentatively suggest that perhaps these
sex-linked hereditary defects, following their typical mode of descent, are
as a group found to be characteristically associated with no other
defects; such would appear to be the case for congenital stationary
night-blindness (with myopia) and haemophilia."

Such a viewpoint, if correct, would have implied the necessity for
formulation of a genetic hypothesis applicable only to human heredity,
i.e. that the human X-chromosome can transmit only one gene for an
abnormality. One would have had to assume that the other genes
present were necessarily normal, or that the presence of more than one
abnormal gene in the X-chromosome constituted a lethal factor, the
association of two sex-linked defects never being observed owing to non-
viability of the resultant organism. In view of the extensive advances
already made in genetical theory by that time, the publication of such a
viewpoint in an authoritative work as late as 1932 on the basis of purely
negative evidence merits some degree of criticism. It is perhaps poetic
justice that the evidence now available for the linkage of such genes in
man should be confined to just those three characteristics—night-
blindness, colour-blindness, haemophilia—which, it was stated in 1932,
ever occurred in association.

Riddell (1937) instanced a pedigree in which haemophilia occurred in
association with colour-blindness: Bell & Haldane (1937) instanced
further cases in which the two defects were found in association. I) 1 one
case of crossing-over was noted, and that was not absolutely certain since
the mother of the case in question may have been a mosaic due to somatic
mutation. The linkage therefore was virtually, if not entirely, complete.
Rath (1938) instanced a further pedigree in which a female known to be
transmitting haemophilia acquired., apparently as the result of mutation, a
gene for colour-blindness and gave rise to four sons, one normal, one
colour-blind., one haemophilic, and one haemophilic and colour-blind,
constituting definite evidence of the possibility of chromosome crossing-
over in man. The data were not sufficient, however, to permit of an
estimate of the frequency of crossing-over between the genes for these
two defects.

In the present work, colour-blindness and congenital stationary night-
blindness with myopia have been found in association in several members of
a pedigree which extends over seven generations. The mode of occur-
Linkage and Crossing-over in Human Sex Chromosomes

renue of the defects necessitates acceptance of the fact that the genes, responsible for these abnormalities must be located in the same X-chromosome and are transmitted as sex-linked recessives. It seems a logical conclusion that the early failure to observe such linkage must be ascribed to the comparative rarity of sex-linked defects, the still greater rarity of occurrence of associated sex-linked defects, and the difficulty of following up human pedigrees.

The present case is complicated by a high frequency of crossing-over of the maternal X-chromosomes, a feature which has resulted in the occurrence in the pedigree of night-blind, myopic, colour-blind males; colour-blind, males; and night-blind, myopic males. It is clear from the subsequent evidence that, in regard to the possibility of linkage of defects and, of chromosomal crossing-over, human heredity very definitely conforms to accepted principles of more general genetics - a fact which is of some scientific importance.

DISCUSSION OF THE PEDIGREE AND OF THE DEFECTS STUDIED

The pedigree covers seven generations and commences with a certain Mr Shurly (I. 2) who, it is almost certain, was night-blind, myopic, and perhaps colour-blind. This deduction, concerning an individual the date of whose birth was probably somewhere around 1760 to 1780, rests upon the fact that his granddaughter, Mrs Emily Clarkson (née Keates) (III. 11), informed her daughter Mrs B-l (IV. 41) that Mr Shurly was blind. The rarity of spectacles until the early nineteenth century, combined with the incidence of bight-blindness and a severe myopia of as much as —14 diopters in the present-day descendants of Mr Shurly, together with the fact that the said individual married and produced several children, renders it highly probable that he was not actually blind but that he did suffer from defects which, in the absence of spectacles and adequate night-time lighting, would have made his capacity for visual perception far less than normal. In the light of this, the information which the -Writer received from IV. 41 is significant and seems to point to Mr Shurly as the earliest known case of the defects in this family.

From the same source (IV. 41) came also the information that Mr Shurly had at least four children of whom one, a daughter, Eliza Mary Ann Shurly (II. 1) (7 : 3 : 1800-1886) married John (Jesse ?) Keates (II. 2) and gave rise to the family under discussion. No trace of the remaining Shurly children has been found, but it is possible that they too may have given rise to descendants exhibiting linked ocular defects.
in the same manner as the members of the present pedigree. The information regarding them is therefore given in view of the possibility that their descendants may be independently located and linked up to the present pedigree.

It seems certain from information given the author regarding John (Jesse?) Keates by IV. 41 and by Mr H. G. B-l. (IV. 15) and Mrs C—e (IV. 16) who are the sole living descendants who remember having met him, that his vision was normal, or at least that he was not night-blind or myopic, a fact which lays the onus for transmission of these defects upon Eliza Shurly (IL 1) and links up with the statement regarding her father's "blindness". Little more is known of these members of generations I and II except that they lived in London, but in the case of generation III it has been possible to check the accuracy of some of the information concerning the members of this generation (all of whom are now dead) by reference to marriage and birth certificates which are available for III. 1 and III. 11 respectively.

Eliza Keates (née Slurly) (II. 1) had six children in the order Eliza Mary Ann Keates (III. 1) (18:12:1828-14:16:1919), Thomas Jesse Keates (III. 4), Alfred Keates (III. 7), Helen Keates (III. 8) (1832-1909), Rachel Keates (III. 10), and Emily Haines Keates (III. 11) (2:5:12:1843-1900). The significance of the pedigree emerges quite clearly on consideration of the descendants of Eliza Mary Ann Keates (III. 1) (Mrs Bull), the eldest of the children. Her husband, Seth Bull (III. 2), was not night-blind or myopic, a fact which has been confirmed by the testimony of several members of the family and from existing photographs, and it is quite certain that Eliza Keates was responsible for the transmission of deuteranopia and also night-blindness with a severe myopia.

The defects appear only in her male descendants, both separately and in association, and the form and extent of their occurrence makes it certain that not only are the defects sex-linked but that they were originally (and are still in some cases) located on the same X-chromosome. It is further obvious from the results that separation of the defects has occurred on several occasions, producing, in addition to the expected night-blind myopic colour-blind males, other males who are colour-blind only or night-blind myopic only—the occurrence of all three types being capable of explanation only if it is assumed that crossing-over of the XX sex-chromosomes can and does occur in the human female.

Throughout this branch of the family the type of colour-blindness found is deuteranopia or green-blindness. Its presence in the affected
individuals has been shown by use of the Ishihara test and has been confirmed independently, on the author's behalf, in the case of several individuals by Mr Rudd, Senior Surgeon of the Birmingham and Midland Eye Hospital, using both Ishihara charts and the Edridge-Greene Lamp. Mr Rudd has also confirmed the myopia and the presence and type of night-blindness in the same individuals and thereby provided an invaluable independent testimony for which the writer is extremely grateful.

The writer has tested as many members of this branch of the family as possible. In the case of the majority of the affected individuals, independent confirmation of the findings and details of the eyesight of the members is available in the form of prescriptions obtained from the oculists who provided them with spectacles, or the specialists originally responsible for drawing up the prescriptions. Details are given in the observational section of this work.

As for the night-blindness found in this branch of the family, it is undoubtedly congenital stationary night-blindness. This follows from the sex-linked mode of its inheritance, the non-progressive nature of the defect (present from earliest recollection but of constant intensity throughout life), and from the complete absence of any signs of fundus degeneration or pigmentation (Mr Rudd's findings). These facts exclude retinitis pigmentosa. The exclusion of any possibility that the defect results from vitamin A deficiency follows from the fact that one of the members, Mr W. W. W—n (V. 24), was given by the "Glaxo" Laboratories, 48,000 international units of vitamin A per day, in the form of "Prepalin" capsules, over a period of eighteen months without any appreciable improvement in his night-blindness. The dosage is some six times that generally accepted as required to clear up a vitamin A-deficiency night-blindness, and the period of treatment much longer than would be required. The author is grateful to the "Glaxo" Laboratories for permission to quote this datum as a confirmation of his findings. The negative result of this experiment may be emphasized here as a warning against the apparently growing tendency in medico-physiological circles to consider all forms of night-blindness as resulting from vitamin deficiency.

There is available, therefore, a variety of independent evidence confirming the writer's observation of the presence of deuteranopia and congenital stationary night-blindness with myopia in the descendants of Eliza Keates 1). She gave rise to three sons (IV. 2. 8. 15) and nine daughters, and two at least of the daughters have abnormal X-
chromosomes produced as the result of crossing-over of the maternal XX-chromosomes—a fact which will be discussed later.

The eldest son, Charles Henry B-1 (IV. 2), was night-blind and severely myopic. His death occurred some years before the writer commenced this work, and it is impossible to say whether or not he was colour-blind. He left no descendants. The next son, Henry Seth B-1 (IV. 8), died in infancy, and there is no information concerning his eyesight. The youngest son, Henry George (IV. 15), was tested by the writer and found to have normal eyesight, and his descendants by his first wife are therefore of no interest to this discussion. His second marriage with his first cousin (IV. 41) who, as will be shown later, was transmitting deuteranopia, was consanguineous. She herself, like all the remaining female members of the pedigree, exhibited none of the defects studied. The description of the occurrence of the defect in the descendants of this marriage is however, best deferred until the branch of the family of which IV. 41 is a member is considered.

Of the nine daughters of Eliza Keates, one (IV. 3) died in infancy, one (IV. 13) died unmarried, and one (IV. 11) although married has no progeny. They need not therefore be discussed further. The remaining six daughters (IV. 4, 6, 9, 16, 18, 20) are all transmitting the ocular defects, the high percentage of female transmitters in this generation being probably the result of chromosome crossing-over. Each has given rise to a group of descendants which must be discussed in some detail.

The eldest of these daughters, Emily Eliza (IV. 4) (Mrs P—s), had one son, Chris. P—s (V. 2), who is night-blind myopic and colour-blind.

The next daughter, Kate Ellen (IV. 6) (Mrs W—t), had three sons, one of whom died in infancy, while his twin is night-blind, myopic and deuteranopic. The third son is not night-blind or myopic, but is in New Zealand and not available for a test of his colour perception.

The third of the six daughters, Alice Clara (IV. 9) (Mrs W—), has given rise to the most important branch of the family. Of her five sons, Clifford Augustus (V. 11) is night-blind myopic and colour-blind as are also his brothers, John Wilfred (V. 15) (the propositus) and William (V. 24). Walter (V. 19) is colour-blind only, while Leslie (V. 22) is night-blind myopic but not deuteranopic. Since the father of these sons (IV. 10) has been tested and found not to be colour-blind he does not enter into the interpretation. It is obvious, considering the mode of occurrence of the three defects in these five sons, in conjunction with its occurrence in other branches of the family, that Alice Clara B-1 (IV. 9) must have carried on one of her sex-chromosomes determinant genes for all three
defects. This chromosome has been passed on intact to three of her sons. In the case of the other two sons, chromosome crossing-over has obviously come into play, causing one of them to be deuteranopic only, and the other to be night-blind myopic but not deuteranopic.

Further, two at least of the five sisters of these brothers are also transmitting the same defects. The eldest sister, Nell (V. 12) (Mrs P—k), has two sons, one of whom, Karl P—k (VI. 13), is night-blind myopic but not colour-blind. His brother, Sydney (VI. 16), has normal eyesight. These members, resident at Toronto, were tested for the writer by Dr J. W. MacArthur of the Toronto University Biology Department, and the writer is extremely grateful for his assistance in this matter.

Three of the remaining four sisters have not produced male issue; but the fourth, Dorothy W—n (V. 25) (Mrs G—y), has one son, Trevor (VI. 27), who is deuteranopic, but not night-blind or myopic, implying a further case of chromosome crossing-over in his or his mother's development. His brother died in infancy, and it is impossible to say whether or not he shared the defect. This section of the family therefore exhibits four indisputable cases of chromosome crossing-over and three cases of normal transmission.

To return to the three remaining daughters of (III. 1). The next, Florence Amelia B—l. (IV. 16) (Mrs C—e), has produced six children including two sons, Harry John (V. 44) and Edgar (V. 46), both of whom are deuteranopic, but not night-blind or myopic, implying a result that can again be due only to chromosome crossing-over, either in their formation or in that of their mother. Their four sisters have given rise to only one son, Douglas S—n (VI. 43), who has normal eyesight.

The next sister, Maud Eugenie (IV. 18) (Mrs J—s), has produced eight sons and six daughters, but it is impossible to say whether any of the latter are transmitters since they are all devoid of male issue. The first of the eight sons, Samuel (V. 55), died in infancy, and the condition of his eyesight is unknown. Three of the sons, Jack (V. 67), Chris (V. 70) and Roy (V. 73), were found to possess normal eyesight, whereas the remaining four, Albert (V. 57), Laurence (V. 64), Harry (V. 68) and Leslie (V. 75), were all night-blind myopic. No trace of deuteranopia was found in this branch of the family, and it seems almost certain that chromosome crossing-over took place in the development of the mother (IV, 18), making it possible for her to transmit only night-blindness and myopia.

Two further cases of crossing-over are found again in the last section of this branch of the family which requires discussion, the descendants of
Emmeline, Maria (IV. 20) (Mrs W—e and later Mrs W—n). Of her three sons, the eldest, Albert W—e (V. 79), is night-blind myopic, but not deuteranopic. The next son, Jack (V. 81), lives in America and has not been tested for deuteranopia but is known to be free from night-blindness and myopia. The third son, Leonard (V. 82), is stated to have had normal eyesight but died of tuberculosis some years ago, and had not been tested for deuteranopia. Of their four sisters, Maud (V. 76) (Mrs V—y) has two sons with normal eyesight. The other married sister, Helen (V. 83) (Mrs M—e), has one son, Brian (VI. 61), who was found to be deuteranopic only, a further case of chromosome crossing-over.

In this branch of the family (the descendants of Eliza Keates (III. 1)) occurs also the only case of colour-blindness which has been introduced into the family from an extra-familial source—that of David W—n (VI. 22), who suffers from incomplete red-green blindness—a form of colour-blindness easily distinguished in the Ishihara test from the green blindness characteristic of other members of the pedigree. Since his father (V. 22) happens to be the one of the five brothers discussed earlier who is night-blind myopic but not colour-blind, VI. 22's colour-blindness obviously derives from his mother (V. 21) who is not in the direct line of the family descent.

This poses an interesting question in that VI. 22 has a sister, Monica W—n (VI_ 23), who will undoubtedly have received from her father (V. 22) one X-chromosome bearing the genes for night-blindness and myopia. She has a 1 in 2 chance of receiving from her mother an X-chromosome bearing a gene for colour-blindness. It is evident from the data given above that crossing-over of the X-chromosomes between the loci of these two genes is very frequent, and it is possible, therefore, that if VI. 23 eventually gives rise to male children, some of them may be found to be night-blind myopic and colour-blind despite the fact that owing to chromosome crossing-over her father's descendants should be free from colour-blindness.

A similar possibility is present in another section of this branch of the family—peculiarly enough, again where crossing-over has caused colour-blindness to be no longer a feature of the inheritance—i.e. in the children of IV. 18. The two brothers, Roy J—s (V. 73) and Leslie (V. 75), have married two sisters (V. 72 and 74) whose father is known to be colour-blind. The potential daughters of V. 75 and V. 74 will receive X-chromosomes with genes for night-blindness and myopia from their father (V. 75) and have a 1 in 2 chance of receiving an X-chromosome with a gene for colour-blindness from their mother (V. 74). The grandsons of V. 75 may
therefore again exhibit night-blindness and myopia in association with colour-blindness. It is a curious reflexion upon the laws of probability that such a situation (i.e. the reintroduction of colour-blindness into the family) should occur in each case in sections of the family from whose inheritance the deuteranopia has been eliminated. This feature, with the fact of the presence of several young unmarried or newly married daughters of night-blind myopic deuteranopic males in the present living generation, should make the history of the next few generations of this family well worth study. For this reason, fuller details of the pedigree than can be reproduced here are being deposited with the Bureau of Human Heredity, 115 Gower Street, London, with a view to the family being available for further study by any future investigators who may care to undertake the task. Pedigrees such as the present one will undoubtedly be rare and the details available will permit of a full follow-up of the present investigation.

It is necessary now to return to consideration of the sections of the pedigree containing the descendants of the remaining children of Eliza Shurly (II. 1). Of these children, Thomas Jesse Keates (III. 4) is stated by Mr S—t (IV. 23), his daughter’s husband, by Mr W. C. W—n (IV. 10), by Mrs W—s (IV. 14) and by Mrs C—e (IV. 16), all of whom knew him well, to have had normal eyesight, with no myopia or night-blindness, and never to have used spectacles. He may of course have been colour-blind, as a result of the chromosome crossing-over which has already been shown to be of frequent occurrence, but there is no evidence on the point. Unfortunately, he has no living descendant in whom colour-blindness could be present as a result of hereditary transmission from him, and the group of his descendants shown in the pedigree chart need not therefore be discussed here.

Alfred Keates (III. 7) is also known not to have suffered from night-blindness or myopia, and again there is no knowledge as to whether or not he was colour-blind. He is reported by IV. 16 and IV. 41 to have married, but it is uncertain whether or not he gave rise to descendants, and for some reason none of the family have been able to provide any further information which would enable a decision as to the point.

Rachel Keates (III. 10) died unmarried at the age of fourteen and therefore needs no discussion. Of the two remaining sisters, Helen. Keates (III. 8) became Mrs Long, and gave rise to one daughter (IV. 28), who cannot be traced, and four sons (IV. 31. 33. 36. 39), none of whom were myopic or night-blind. Information is available regarding the numbers of their descendants, but such of them as can be traced are free
from ocular defects and it is probable that Helen Keates 8) was not
transmitting the defects characterizing the family. There is no evidence
that she transmitted colour-blindness, but it has not been possible to
locate and test sufficient of her descendants to exclude definitely such a
possibility. Again, further discussion of this section of the pedigree would
serve no useful purpose.

In the case of the remaining sister, Emily Keates (M. 11) (Mrs
Clarkson), the circumstances are different. Of her four sons (IV. 40. 42. 44.
48), two (IV. 40. 48) died in infancy. Charles Alfred (IV. 44) was tested
by the writer and was found to have normal eyesight. His brother, Samuel
Henry Thomas (IV. 42), died of enteric fever while fighting in the Boer War
and is stated by IV. 41 and IV. 44 to have had normal eyesight, although it is
again impossible to state definitely that he was not colour-blind.

Two of Emily Keates's daughters have married, one, Emily Ann
Eliza (IV. 41), having married her first cousin, Mr H. G. B-l. (IV. 15),
and given rise to two daughters and two sons (V. 37. 40. 41. 42), the
younger of the latter being green-blind, but not night-blind or myopic.
The defect may have been inherited from his grandfather, Mr Clarkson (III. 12),
of whom; being dead, it is impossible to say he was not deuteranopic, but—
in view of the frequency of occurrence of deuteranopia the descendants of
his grandmother's sister it seems fairly certain that Emily Keates 11) was
transmitting deuteranopia received via Eliza, Shurly from Mr
Shurly. If this conclusion be correct, it implies that in formation of the
ovum which gave rise to Emily Keates (III. 11) or her daughter Emily C—
n (IV. 41), crossing-over of the. XX-chromosomes occurred, allowing of
the transmission of deuteranopia unaccompanied by night-blindness and
myopia. This conclusion seems hardly disputable in the light of the
evidence already given earlier concerning the descendants of Eliza Keates
(III. 1).

The remaining daughter of Emily Keates, i.e. Kate Clara Alice
(IV. 46), has produced one son and a daughter. Her son is not night-
blind or myopic but may be colour-blind. This, however, cannot be
determined, since he is at present in Vancouver and is not available for
test.

Since Prof. J. B. S. Haldane, F.R.S., has very kindly consented to
provide a statistical appendix to this work giving a fuller mathematical
treatment of the linkage and cross-over ratios than the writer is capable of
providing, it is necessary here to add only a few remarks concerning this
aspect of the pedigree.
414 Linkage and Crossing-over in Human Sex Chromosomes

The pedigree contains 273 individuals of whom 183 are in the direct line of descent from Mr Shurly (I. 2). Of these latter, forty-five are deceased, and twenty-eight owing to emigration were not available for test. Of the remaining 110 members in the direct line of descent, seventy-one (i.e., 64.6%) have been interviewed and tested by the writer or by other competent persons on his behalf, the majority of the untested members available in this country being of little, if any, interest to the study (e.g., females with no male issue). In all, ninety-nine members of the pedigree have been tested, most of the twenty-eight extra-familial members having been tested to ensure that they were not responsible for introducing any of the defects from an extra-familial source.

Of the 183 direct members of the family, eighty-seven are males and ninety-six females. Thirteen of these latter have produced sons or grandsons exhibiting the defects and are thus known to be transmitters. A further nine—daughters of males with the defects—will, in the normal course of events, be found to be transmitters when they marry and produce progeny. The ninety-six females therefore include at least twenty-two transmitters (i.e., 22.9%), while of the eighty-seven males, and their forerunner (I. 2), twenty exhibit the ocular defects (i.e., 22.7%), a surprisingly close correspondence. Of these twenty males, five are night-blind, myopic, deuteranopic; seven are night-blind, myopic but not deuteranopic; six are deuteranopic only; a further two (both dead) are known to be night-blind myopic, while data concerning their perception of colour is not available.

The thirteen females producing sons or grandsons with the defects have produced thirty-eight sons, of whom nineteen (Mr Shurly I. 2 is excluded here) are so affected, but thirteen of the thirty-eight sons, owing to death or emigration, cannot be said to be free from deuteranopia and have to be disregarded. The defects therefore are exhibited by 76% of the sons for whom data is available, the 26% excess over the expected 50% being due probably to the lack of complete data, and to the high degree of chromosome crossing-over.

Of the forty-two daughters of the thirteen transmitting females, twelve have produced progeny with the defects, but only three can be said with some certainty to be non-transmitters, seven having not married, nine having died unmarried, while twenty-three have no male issue, or have produced too few sons to permit of a decision as to whether or not they are transmitting.

If II. 1 be assumed to be transmitting the genes for both colour blindness (c) and night-blindness-myopia (n) on the same X-chromosome,
then the cross-over ratio between these two genes can be determined empirically within certain limits by consideration of Tables I and II.

This assumption, as can be seen from consideration of Prof. Haldane's statistical appendix, is not necessarily correct, since II. 1 may have the genetic constitution \(\frac{c+}{c+n}\) (if both defects are derived from her father), \(\frac{c+}{+n}\) (if the colour-blindness comes from her mother and night-blindness from the father), or \(\frac{++}{+n}\) (if the colour-blindness is introduced by her

**TABLE I**

_Giving the lowest possible value for the cross-over ratio between the genes c and n_

<table>
<thead>
<tr>
<th>Daughters</th>
<th>Transmitting night-blindness, myopia, deuteranopia (i.e. non-cross-overs)</th>
<th>Transmitting night-blindness and myopia, or only deuteranopia (i.e. cross-overs)</th>
<th>Sons</th>
<th>Night-blind, myopic, and deuteranopic (i.e. non-cross-overs)</th>
<th>Night-blind, myopic, or only deuteranopic (i.e. cross-overs)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mother</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>II. 1</td>
<td>III. 1</td>
<td>III. 11</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>III. 1</td>
<td>IV. 4</td>
<td>IV. 16</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>IV. 6</td>
<td>IV. 18</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>IV. 9</td>
<td>IV. 20</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>III. 11</td>
<td>IV. 41</td>
<td>V. 2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IV. 4</td>
<td>V. 27</td>
<td>V. 12</td>
<td></td>
<td>V. 11</td>
<td>V. 19</td>
</tr>
<tr>
<td></td>
<td>V. 15</td>
<td>V. 24</td>
<td></td>
<td>V. 15</td>
<td>V. 22</td>
</tr>
<tr>
<td>IV. 20</td>
<td>V. 83</td>
<td>V. 24</td>
<td></td>
<td>V. 24</td>
<td>V. 22</td>
</tr>
<tr>
<td>IV. 41</td>
<td>V. 79</td>
<td>V. 22</td>
<td></td>
<td></td>
<td>VI. 27</td>
</tr>
<tr>
<td>V. 27</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>VI. 61</td>
</tr>
<tr>
<td>V. 83</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

husband (II. 2)). It seems most probable, however, that II. 1 had the genetic constitution \(\frac{c+}{c+n}\), and although the cross-over ratio, assuming this condition, is found to be lower than that obtained later by Prof. Haldane's more detailed analysis, it is still of the order of 43%, a significantly high value.

In Table I III. 11, V. 25, and V. 83 are presumed to be capable of transmitting all the defects. In consequence, ten females have produced nine females and five males normally, and three females and six males
by processes involving cross-aver of the XX-chromosomes. The crossover ratio is therefore 39.1% (nine of twenty-three children).

Table IT permits deduction of the upper limit of the cross-over ratio by assuming crossing-over to have occurred in the development of III. 11, V. 25 and V. 83 instead of in that of their sons or of their daughters or granddaughters who are transmitting the defects.

### TABLE II
*Giving the highest possible value for the cross-over ratio between the genes c and n*

<table>
<thead>
<tr>
<th>Mothers</th>
<th>Transmitting night-blindness, myopia, deuteranopia (i.e. non-cross-overs)</th>
<th>Transmitting night-blindness, myopia, or only deuteranopia (i.e. cross-overs)</th>
<th>Sons</th>
</tr>
</thead>
<tbody>
<tr>
<td>II. 1</td>
<td>III. 1</td>
<td>IV. 4</td>
<td>V. 2</td>
</tr>
<tr>
<td>III. 1</td>
<td>IV. 4</td>
<td>IV. 16</td>
<td>V. 19</td>
</tr>
<tr>
<td></td>
<td>IV. 6</td>
<td>IV. 18</td>
<td>V. 22</td>
</tr>
<tr>
<td></td>
<td>IV. 9</td>
<td>V. 12</td>
<td>V. 24</td>
</tr>
<tr>
<td></td>
<td>IV. 20</td>
<td>V. 27</td>
<td>V. 79</td>
</tr>
</tbody>
</table>

In this case six females have produced five females and five males normally, and six females and three males by processes involving chromosome cross-over. The cross-over ratio is therefore 47.4% (nine of nineteen children).

This admittedly empirical treatment indicates the cross-over ratio to be within the extreme limits 39.1-47.4%, or, taking the mean of these values, to be $43.25 \pm 4.15\%$. If this value which, for reasons given above, is lower than that (64.8%) given by proper statistical treatment be taken as correct, 71.7% of the sons of the transmitting females should exhibit the defects instead of the expected 50%, a fair agreement with the observed 76% given earlier. Further, it follows that the human X-chromosome map must be a long one, as in *Drosophila* spp. and *Gallus*, and not short as in *Lebistes*. Taking the length of the human X-chromosome as 100 units, the present work therefore indicates the distance between the gene for deuteranopia, and that (or those) for night-blindness and myopia, to be some $43.25 \pm 4.15$ units. Since, in no case, have any of these defects been transmitted by an affected male to his sons, it
can be said also with some certainty that the genes for deuteranopia and night-blindness with myopia are located on the non-pairing segment of the X-chromosome, i.e. the section which does not cross-over with the male Y-chromosome (a behaviour which Haldane (1936) states is possible). It cannot be said whether or not the night-blindness and its accompanying myopia are both governed by the same gene, for in no case has the one defect been found without the other. Myopia does occur in certain males in the family without an accompanying night-blindness or deuteranopia, but in all such cases the degree of the myopia is very small (generally ca. 1 diopter), and in most its occurrence can be traced to an extra-familial source. There is no evidence of any crossing-over between the genes for night-blindness and myopia, nor is there any evidence in any other pedigree known to the writer, and it can be stated with certainty that these two defects are either governed by the same gene, or by two genes in positions so closely adjacent that the cross-over ratio between them is very small. In consequence of this it has been possible to simplify discussion of the crossing-over in this work by assuming only two genes to be concerned—one determining deuteranopia and $43.25 \pm 4.15$ units distant from that determining the occurrence of night-blindness with myopia.

As for the defects themselves, it will be seen from consideration of the details in the observational section that their intensity varies to some extent. No quantitative estimation of the degree of night-blindness in any of the individuals has been possible owing to the non-availability of apparatus suitable for such estimation. It is believed, however, that the capacity for receiving weak light impulses is a function of the peripheral rods of the retina, and in this respect certain findings made by Mr Rudd are important. In his tests on V. 4. 11. 15. 22, Mr Rudd noted, in three of the four cases, a general contraction of the fields of vision, which contraction can be observed in the Perimeter and Scotometer Charts illustrated in the observational section of this work. This contraction was scarcely noticeable in the case of V. 4, whose night-blindness, however, is not as severe as in the other three cases. A similar contraction was observed by Dr J. X. Robert of Toronto in the case of VI. 13.

In the case of all the night-blind, individuals seen by the writer, it was stated that they had suffered from the defect from their earliest recollection, and that its intensity had always seemed approximately the same. V. 11 and V. 24, however, both stated that the night-blindness appeared less troublesome when they felt in a really good state of health,
but other members failed to confirm this impression. Several of the affected individuals stated also that they "could not have too much light", and V. 11 noted, while on a Mediterranean cruise, that on occasions when other passengers had to use dark spectacles because of the intensity of the sunlight, he himself had never before experienced such a high degree of visual acuity. It seems clear, therefore, that even in the presence of what would constitute an excessive source of light to a normal individual, the night-blind person still cannot perceive the light intensity at its fall value. This feature does not appear to have been noted by previous observers and, is significant in its implication that the perception of light intensity by a night-blind person does not at any time attain the capacity for perception of light intensity of a normal-sighted individual—this applying not only when the light source is weak but also when it is excessive. If anything, these observations agree with the theory that light perception is a function of the peripheral rods and the visual purple pigment, and that hereditary night-blindness is due to a congenital deficiency in the amount of visual purple pigment available in the retina. It has already been pointed out that this hereditary deficiency cannot be compensated by vitamin A treatment, and it is possible that the pigment deficiency is physiologically due to a congenital incapacity to utilize vitamin A in the production of visual purple.

The degree of myopia present in the affected individuals is also subject to some fluctuation, varying from -2 diopters in V. 4, -6 diopters in V. 24, -7 in V. 75, to -9.5- -14 in V. 15. 22. 57. 68. There is some evidence, too, that the degree of the defect becomes greater as the affected individual grows older—Contrary to the general principle that the focal length of the eye increased with age—for V. 57 exhibited a myopia of -8 diopters in 1898, and -9.5 in 1930; V. 15 had a myopia -8.5 in 1931, and -10.5 in 1937; while V. 68 had a myopia of -12 in 1921, and -14 in 1933. Such comparisons, unfortunately, are not available for more of the affected individuals.

The degree of deuteranopia shows somewhat less variation, the majority of the affected males being quite distinctly green-blind. In a few cases, however, the individuals on being tested with the Ishihara, charts were able on tests 22-25 to read the numbers which the green-blind individuals cannot generally distinguish, although in all such cases the individuals took some time to read the numbers distinctly, and stated that they were much less distinct than the accompanying figures, which are easily visible to the deuteranopic. V. 2, however, proved somewhat abnormal in being apparently totally colour-blind, although
this may perhaps be due to the effects of an accompanying nystagmus. There is distinct evidence therefore that the actual degree of intensity of the defects varies considerably in individuals who are undoubtedly deriving their genes for these defects from the same ultimate source. Whether or not this variation is due to slight alterations in the gene structures during their transmission it is impossible to say. It is clear, however, that such variation, together with the elimination of one or other of the defects in some of the members as a result of chromosome crossing-over, shows that evolutionary processes are by no means at a standstill in the human species. Under more unfavourable environmental conditions such changes might well affect the survival value of an individual.

In most but not all of the cases the night-blindness and myopia were found to be accompanied by marked astigmatism, and one individual also exhibited cataract, and two others nystagmus. The normal members of the family were surprisingly free from such alternative eye defects except in one or two small groups.

OBSERVATIONAL

The following relevant data were acquired concerning the affected individuals, their colour perception being determined by means of the Ishihara charts involving the reading of twenty-five test numbers. Normal-sighted individuals give readings different from those given by deuteranopes, the two sets of readings being as follows:

<table>
<thead>
<tr>
<th>Normal</th>
<th>Deuteranope</th>
<th>Normal</th>
<th>Deuteranope</th>
</tr>
</thead>
<tbody>
<tr>
<td>12</td>
<td>13</td>
<td>5</td>
<td>-</td>
</tr>
<tr>
<td>8</td>
<td>8</td>
<td>7</td>
<td>-</td>
</tr>
<tr>
<td>6</td>
<td>5</td>
<td>16</td>
<td>-</td>
</tr>
<tr>
<td>29</td>
<td>10</td>
<td>73</td>
<td>-</td>
</tr>
<tr>
<td>57</td>
<td>35</td>
<td>-</td>
<td>5</td>
</tr>
<tr>
<td>3</td>
<td>2</td>
<td>-</td>
<td>2</td>
</tr>
<tr>
<td>6</td>
<td>5</td>
<td>-</td>
<td>45</td>
</tr>
<tr>
<td>15</td>
<td>17</td>
<td>-</td>
<td>73</td>
</tr>
<tr>
<td>74</td>
<td>21</td>
<td>26</td>
<td>2</td>
</tr>
<tr>
<td>2</td>
<td>-</td>
<td>42</td>
<td>4</td>
</tr>
<tr>
<td>6</td>
<td>-</td>
<td>35</td>
<td>3</td>
</tr>
<tr>
<td>97</td>
<td>-</td>
<td>90</td>
<td>9</td>
</tr>
<tr>
<td>45</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

In the observational details, readings signifying deuteranopia are underlined:

I. 2, Mr Shurly, late eighteenth century. Stated by his great-granddaughter (IV. 41), on information from her mother (III. 11), to have "been blind"—probably night-blind myopic.

IV. 2. Charles Henry Bull (5. 12. 1852-1916). Hairdresser, Birming-
ham. Stated by his sisters (IV. 16 and IV. 18), his brother (IV. 15), his nephew (V. 15), his brother-in-law (IV. 10), to have been night-blind and, severely myopic. The latter recollects "piloting him by the arm through the streets after dark". There is no data concerning his colour perception.

V. 2. Chris P-s. University lecturer, Brussels. Tested January 1940 for the author by P. I. Wilson, B.Sc. Night-blind, myopic and totally colour-blind, the Ishihara charts appearing to him as a “blur” in which the coloured numbers could not be differentiated. M. Copper (optician) of Brussels gave the following data regarding his vision when tested in 1935: R. -7 sph., -0.75 cyl., 75° ax.; L. 7 sph., -0.75 cyl., 75° ax.

V. 4. Norman W-t (1883-). Architect's designer, Birmingham. Night-blind, myopic and incomplete deuteranopic. His readings in the Ishihara test were: 12, 8, 6, 79, 37, -5, 17, 71, -5, 87, -2, -7, -2, -2, 2, 4, (2), 3 (5), 3 (6). Tested in July 1939. Aet. 56. The brackets in tests 23-25 indicate the reading of the enclosed figure to have been delayed and the underlined tests are those indicating red-green or green-blindness. Mr Harrison (optician) of Birmingham gave the following data regarding his eyesight when tested in 1937: R. -2 sph., -3.75 cyl., 120° ax.; L. -1.75 sph., -3.75 cyl., 40° ax.; Va =6/18. He was examined for the writer by Mr Rudd, Senior Surgeon of the Birmingham and Midland Eye Hospital, on 26 July 1939, who stated: "History of Night-blindness - vision with correcting lenses R. 6/12, L. 6/9; incomplete red-green blindness - Fundi show no evidence of Retinitis pigmentosa - Fields of vision full."

Figs. 1 and 2 are the perimeter and scotometer chart readings for the right and left eyes respectively of V. 4 as provided by Mr Rudd.

V. II Clifford Augustus W-n (1881-). Pianist, Birmingham. Night-blind, myopic and deuteranopic - the myopia ca. -13. He was tested with Ishihara charts on 7 July and again in August 1939, giving the same readings each time: 12, 6, 70, -5, 17, 71, (2), (6), -2, -2, -2, 2, 4, 3, 9. Mr Wood-White (Late Senior Surgeon of the Birmingham and Midland Eye Hospital) tested him on 24 April. 1917 stating "very high myopia with consequent nystagmus and night-blindness. R. 6/24 (corr.), L. less than 6/60 (corr.)." Mr Rudd tested him on 2 August 1939 and stated "History of difficulty in getting about in dim light - vision with correcting lenses R. 4/12, L. 6/60 - Fields of vision: moderate general contraction - Fundi: extensive myopic degeneration-Nystagmus - appears to be an incomplete red-green blind."
Figs. 3 and 4 give the perimeter and scotometer chart readings provided by Mr Rudd.


12, 3, 5, 70, 35, 2, 5, 17, 21, , , , , , , , 5, 2, 45, 73, 2, 4, 3, 9.

Mt Harrison (optician), Birmingham, tested him in 1931 and found

R. -8.5 sph., -1.75 cyl.; Va 8/18; L. -8.5 sph., -2.5 cyl.; Va 8/18.

He was tested again on 12. September 1937 giving R. -10.5 sph., -2.0 cyl., 90° ax.; Va 8/24; L. -8.0 sph., -3.0 cyl., 90° ax.; Va 8/24. Mr Rudd tested him on 26 July 1939 and stated: "History of night-blindness—Vision with correcting lenses R. 6/18, L. 6/18—Fields of vision: some general oontra—Fundi: myopic crescents—incomplete red-green blind." Figs. 5 and 6 are the relevant perimeter and scotometer charts.

V. 19. Walter W—n (20. 11. 1890—). Manufacturer, Birmingham. Deuteranopic only. Tested in July 1939 giving the Ishihara readings:

12, 3, 5, 70, 35, 2, 3, 17, 21, , , , , , , , 5, 2, 45, 73, 2, 4, (2), 3, (5), 9. He showed no evidence of night-blindness or myopia.

V. 22. Leslie W—n (1897-) Manufacturer, Birmingham. Night-blind, myopic but not colour-blind. He was tested in July 1939 and gave quite normal Ishihara readings. Mr Harrison (optician), Birmingham, examined him on 3 June 1937 and found.

R. -12 sph., -0.75 cyl., 60° ax. ; L. -11, and stated cataract and iritis to be present also. Mr Rudd examined him on 2 August 1939 and stated: "History of defective vision especially in dim light—Vision with correcting lenses R. 6/18, L. 6/18—Fundi: some myopic degeneration round, the discus—Fields of vision: slight nasal contraction only : Colour vision normal." Figs. 7 and 8 are the relevant perimeter and scotometer charts.


12, 3, 5, 70, 35, 2, 5, 17, 21, , , , , , , , 5, 2, 45, 73, 2, 4, 3, 9. His optician's report gave R. -6.5 sph., L. -6.75 sph., -0.75 cyl., 80° ax. : the date of the test being 26 January 1939. He was given 48,000 units of vitamin A per day for 18 months by the " Glaxo " Laboratories without any noticeable improvement in his night-blindness.

V. 42. Frank Edward. B-l (1917). Army, Torquay. Deuteranopic according to his parents and, several of his cousins. Owing to the war he was not available for testing. No night-blindness or myopia.
422 Linkage and Ct088i941-0VC1 in Human Sex Chromosomes


26. 7. 39. $\frac{1}{2}^\circ$ while obj.

2. 8. 39. $2^\circ$ white obj.

26. 7. 39. $\frac{1}{2}^\circ$ white obj.
V. 44 Harry John C—e. Air Force, Birmingham. Deuteranopic according to the R.A.F. report, but passed for the, ground staff. Owing to the war he again was not available for testing, but his wife states he cannot distinguish green from blue, or purple from red, and the author, visiting him at night when the Ishihara charts could not be utilized, verified this statement. No night-blindness or myopia.

V. 46. Edgar C—e. Mechanic, Coventry. Deuteranopic. Tested on 6 January 1910, he gave the Ishihara readings: 12, 3, 5, 70, 35, 2, 5, 17, 21, 2, 2, 2, 2, 2, 5, 45, 73, 2, 4, 3, 9. No night-blindness or myopia.

7 January 1940, he gave normal Ishihara readings. Mr Lloyd-Owen of the Birmingham and Midland Eye Hospital tested him an 29 April 1898, giving R. –8 sph., L. –8 sph. Mr Charnley and Sons (opticians), Leamington Spa, tested him on 2 January 1930, giving R. -9.5 sph., L. –9.5 sph.

V. 64. Lawrence J—s (10. 7. 1899–). Gardener, Par. Night-blind, myopic, hut not colour-blind. Tested on 1 December 1939, he gave normal Ishihara readings. Mr Huddy (optician), St Austell, gave the following data for his eyesight: R. -3.5 sph., -1.5 cyl., 100° ax.; L. –5.5 sph, –1.5 cyl., 80° ax.

V. 68. Harry J—s (24. 11. 1903-) Gardener, Par. Night-blind, myopic, but not colour-blind. Tested on 1 December 1939, he gave normal Ishihara readings. Mr East, of the Royal Cornwall Infirmary, tested him on 23 August 1924 and found R. –12 sph., L. –11 sph. Mr Rolston (optician), Plymouth, tested him on 6 October 1933 and found R. –14 sph., L. –14 sph.

V. 75. Leslie J—s (29. 2. 1912–). Gardener, St Austell. Night-blind, myopic, but not colour-blind. Tested on 2 December 1939, he gave normal Ishihara readings. Mr Hodge (optician), Truro, tested him on 23 November 1933 and found R. –7 sph., –3 cyl.; L. –7 sph., -3 cyl.

V. 79. Albert W—e. Administration, Singapore. Information regarding V. 79 has been provided by Dr A. D. Williamson, F.R.O.S., Ophthalmic Surgeon and Physician, General Hospital, Singapore, who tested him for the author on 26 January 1940, and states: "I have examined Mr A. E. W—e to-day. Eyes are normal externally; pupils equal and react normally; ocular movements full. Visual acuity: R. –11.5 sph. -2 cyl. 123° ax. = 6/12 partly. L. – 12.0 sph. – 2.0 cyl. 65° ax. 6/12 partly. Fundi show no abnormality beyond slight myopic crescents. With the ophthalmoscope there is detected a slight constant horizontal oscillatory nystagmus. Tested by the Ishihara plates there is no defect whatever of colour vision. A marked degree of hemeralopia is present."

V. 79 is therefore night-blind, myopic but not colour-blind—an example of crossing-over.

V. 13. Karl P—k (1914). Research Chemist, Toronto. Tested for the writer in December 1939 by Dr J. W. MacArthur of Toronto University. Night-blind, myopic, but not colour-blind. Dr J. N. Robert of Toronto stated of him: "This patient has been under my care since March 1932 for the care of his eyes. At that time vision was as follows: R. –9 sph.,
-2 cyl., 105° ax.; Va 6/36; L. -6.5 sph., -2.5 cyl., 75° ax.; Va 6/36. Horizontal nystagmus when looking to extreme right or left. Examination of fundi showed discs inverted. Night-blindness was complained of at that time. Most recent examination 15 January 1940 showed no change in vision or refraction. Night-blindness and nystagmus continued as above. The field of vision is as shown on the enclosed chart" (i.e. Figs. 9, 10).

V. 22. David W—n (1924-). Birmingham. Incomplete red-green blind - the defect probably inherited from his mother. Tested in July 1939, he gave the Ishihara readings: 12, 8, 5, 70, 57, 2, 5, 17, 21, z, z, z, z, z, z, z, z, 5, 45, 73, 28, 42, 85, 96.

VI. 26. Trevor G—y (1929-). Birmingham, Deuteranopic, but not night-blind or myopic. Tested in July 1939, he gave the Ishihara readings: 12, 8, 6, 70, 85, 2, 5, 17, 21, z, z, z, z, z, z, z, z, z, 5, 2, 41, 21, 2, 4, 3, 9.

VI. 61. Brian M—e (1932-). Birmingham. Deuteranopic, but not night-blind or myopic. Tested in July 1939, he gave the Ishihara readings: 12, 3, 5, 70, 53, 2, 5, 11, 21, z, z, z, z, z, z, z, z, z, 5, 8, 15, 73, 3, 4, 3, 9.

COMPILATION OF THE DATA

The technique of accumulating facts concerning the inheritance of human characteristics necessarily differs from case to case and, in fact, success depends not so much upon the capacities of the investigator as upon the attitude of the family concerned toward the research, and on the extent to which its members have maintained contact with one another. Given full cooperation by the family, the investigator has still to face up to the task of unearthing details long since submerged in the subconscious recollection and, worse still, has often to trace long-lost relatives with only a meagre amount of information to commence with. It is impossible to follow a standard procedure under such circumstances - much depends on the investigator's intuition - but an account of the manner in which the information given in the present work was obtained may be of some utility.

The present work commenced as the result of a visit by the writer to the home of Mr Paul I. W—n (VI. 18) during Whitsun 1939 - a visit resulting from the fact that both the writer and VI. 18 were engaged upon chemical research in the same laboratory at the University of Birmingham. During the course of conversation, Mr J. W. W—n (VI. 15), the father of VI. 18, mentioned that he suffered from night-blindness -
the accompanying severe myopia being an evident feature. While dis-
cussing possible physiological causes of the defect with the writer, V. 15 stated that it was present also in three of his four brothers and in several more distant relatives, all males. The compilation of a rough pedigree which followed this statement soon made it evident that the defect was sex-
linked, and the writer, having some knowledge of genetics, decided to follow up the pedigree somewhat further than was possible with the information provided by V. 15 alone.

A further more significant detail soon appeared in consequence of a purely chance remark made by V. 15, who, referring to his work as an architect, stated that despite his myopia and night-blindness he had an excellent perception of colour tints and went on to remark: In fact—what my wife says is blue very often isn't really blue at all."

Knowing that colour-blindness is sex-linked and is more frequent in males than in females, and feeling that Bell's (1932) statements regarding the non-association of sex-linked defects were founded on somewhat meagre evidence, the writer—a few days later—tested V. 15 for colour-blindness by means of the Ishihara charts and found that he was indisputably green-
blind or deuteranopic. This discovery of the presence of sex-linked night-
blindness and myopia together with deuteranopia made it imperative that the family pedigree should be followed up. Mr Paul I. W—n (VI. 18) readily agreed to help in the locating and visiting of such of his relatives as were available in the Birmingham district, and, in providing introductions to them and accompanying the writer on these visits, rendered invaluable service which alone made this research possible. In consequence of this help provided by VI. 18 all those members of the family who had to be seen co-operated readily—a feature which immediately overcame the major difficulty of this type of research, that of persuading members of a family to allow an investigator to enter their homes and ask numerous, sometimes perhaps unwelcome, questions.

Mr W. Clifford W—n (IV. 10) was first visited, in order to obtain the addresses of members of the family whose location was unknown to V. 15 and VI. 18 and also to obtain data regarding the deceased members of earlier generations—a visit which resulted in the construction of a pedigree chart smaller than that illustrated, but containing most of its major details. The details of the chart were then filled in, checked and continually counter-checked, as the result of visits by the investigator to members of each of the branches of the family. All the information obtained was checked as carefully as possible by reference to documentary evidence (e.g. family bibles, marriage and birth certificates, census
records) and it can be said of the pedigree chart as illustrated, that the majority of its details, e.g. orders of birth, numbers of children, etc., are beyond dispute.

Having obtained this preliminary idea of the extent of the family, the writer commenced a systematic series of visits to as many of its members as possible—testing them all for colour-blindness and, where other eye defects were present, obtaining proper data concerning these from the oculists and specialists who had examined the eyes of the affected members. It is impossible here to specify the various ophthalmologists who thus aided the work, but the writer is more than grateful for the independent evidence they thus provided. On no occasion did the writer meet with any difficulty in obtaining their co-operation.

In the absence of any simple scientific apparatus for testing night-blindness it was necessary to rely upon statements from affected members as to whether or not they suffered from this defect. The ever-accompanying severe myopia, generally —10 to —14 diopters, made it easy, however, to locate such affected members, and in most of the cases it was necessary only to observe their behaviour under twilight conditions in order to confirm the presence of the night-blindness. Even in well-lit streets at night-time, such affected members had to be led by a normal person—a feature which the writer confirmed in most of the cases. In the cases of V. 4, V. 11, V. 15 and V. 22 the affected members were further tested for the writer by Mr Rudd, Senior Surgeon of the Birmingham and Midland Eye Hospital, as stated earlier. These tests confirmed the presence of night-blindness and severe myopia in all four cases, and also the presence of deuteranopia in the first three cases and its absence in V. 22. The deuteranopia in V. 4, 11 and 15 was confirmed by Mr Rudd by means of the Ishihara test, and they were also tested, with the Edridge-Green lamp. The use of anything other than the largest apertures of this latter apparatus was, however, made impossible by the high myopia of the cases, and under such restricted conditions of test they were, found to be capable of distinguishing between green and blue but not between red and purple, both of which were called red. The writer later found that several of the deuteranopic members confused not only green and blues but also red and purple, one of them (V. 46) confessing to a complete inability to recognize the latter colour.

So much for the manner in which, proper confirmation was obtained concerning the presence or absence of the defects in various members of the family. The investigation proper commenced with visits to V. 11, V. 19 and V. 22, three of the four brothers of V. 15, the propositus. Of
these, V. 11 was found to be deuteranopic, night-blind and myopic. V. 19 was found to be deuteranopic only, and V. 22 was found to be night-blind myopic but not deuteranopic. It was immediately obvious that IV. 9—the mother of these four brothers—was capable of transmitting night-blindness, myopia and deuteranopia all on the same chromosome, and that to account for her two sons (V. 19 and V. 22) being only deuteranopic and night-blind myopic respectively, it was necessary to assume that crossing-over of the maternal XX sex-chromosomes had occurred during the process of formation of these two individuals. The real significance of the present work thus emerged at its very beginning.

The investigation was gradually extended—until finally the writer had tested the majority of the members available in the Birmingham area, and several other cases of the occurrence of the defects were located, several members being discovered who were deuteranopic only and several who were night-blind myopic and deuteranopic.

In October 1939, the writer commenced certain other researches in London and was thus enabled to test V. 21, who was again found to have all three eye defects. Data for the descendants of V. 12 was obtained for the writer by Dr J. W. MacArthur of Toronto University, whose assistance in this respect was invaluable. The data regarding the descendants of IV. 15, IV. 18, III. 4 and III. 8 were, however, still incomplete, and the author was fortunately enabled to make several long journeys, to obtain the necessary data, by the Medical Research Council who financed the said journeys—an action for which the writer is extremely grateful. In the ease of the descendants of IV. 15 and IV. 18, adequate data was obtained and IV. 41, the wife of IV. 15, provided the information regarding Mr Shurly (I. 2), which was discussed earlier and was of importance in determining the source of the defects in the first few generations.

Most of the descendants of III. 1 were unfortunately found to have emigrated to America, and it was not found possible to locate their whereabouts or to determine their numbers with exactitude. In the case of the descendant of III. 8, the sole information available originally was a statement by IV. 16 that one of the sons, Walter Long (IV. 33), had worked on St Katherine's Dock, London, some sixty years ago. Further details of this branch' of the family were lacking. A visit to St Katherine's Dock succeeded, after some tribulation, in bringing to light a foreman who had known IV. 33, and his information, followed by reference to the offices of the Port of London Authorities, provided details as to the date of death of IV. 33 (1931) and his address at that time.
His wife (IV. 34) was found, on enquiry, to have left the said address, but by dint of much enquiry among her late neighbours, her present whereabouts were located and she was visited. In consequence it was found possible to locate and visit some of the children of IV. 33 and of his brother (IV. 39) and also to determine, with some reliability, the numbers of descendants of III. 8. Unfortunately, however, this section of the family appears to have lost touch with its immediate relatives, and it was not found possible to follow up its members as adequately as the rest of the pedigree. It can however, be said with confidence as a result of what information was available, that this section of the family exhibits no night-blindness or myopia. If deuteranopia were present it would represent only one further chromosome cross-over, in the development of III. 8, so that the incompleteness of the information does not constitute a radical deficiency in the pedigree.

In the main, therefore, the pedigree is complete except that, as indicated earlier, full information is not available for two affected members who are known to have been night-blind myopic, but, being dead, cannot be stated to have been deuteranopic. A few other males who are dead or have emigrated may possibly be, or have been, deuteranopic, but the fact cannot be determined. It is certain, however, that all these male members of the pedigree who have been afflicted with night-blindness and myopia have been listed. The little extent to which the data is incomplete makes no difference to the fundamental conclusions of the work.