Experientia 42 (1986), Birkhäuser Verlag, CH-4010 Basel/Switzerland

Some trends in medical populations genetics**

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** Received for publication 11 May 1984

Summary. Five topics concerning medical population genetics have been selected for discussion: in the field of population cytogenetics, the frequency of chromosomal aberrations and the roles of mutation and selection in the maintenance of balanced rearrangements are studied; the long term genetic effects of treatment and prevention of genetic diseases are reviewed; the relationships between malaria and the sickle-cell trait are discussed; some recent works concerning human DNA polymorphisms in the field of population genetics are presented, and finally, some methods of genetic epidemiology are described.

Key words. Cytogenetics; chromosomal aberrations; genetic diseases; malaria; sickle-cell trait; DNA polymorphisms; genetic epidemiology.

Population genetics deals with the genetic structure of natural populations and is therefore concerned with the evolutionary forces of selection, mutation, drift and migration. In 1908, Hardy and Weinberg independently set out the fundamental theorem of population genetics; 1908 can be considered as the year of birth of this branch of genetics. The mathematical models underlying population genetics were first extensively formulated between 1920 and 1940 by Fisher, Haldane and Wright and later by many others; among them Malecot, Kimura and Crow should be cited. Population genetics has not only exercised a profound influence on the thinking of evolutionists but also found important applications in human and medical genetics.

Much work in recent years has dealt with the genetics of 'intellectual aptitudes' despite the difficulties of using genetic methods of analysis in this field[48]. Particularly, Feldman and Lewontin[23] and Jacquard[31] have shown the difficulties and the virtual impossibility of using a parameter such as heritability in human genetics.

In this paper, we shall review some topics concerning medical population genetics.

Population cytogenetics

During the last fifteen years, it has become evident that chromosome abnormalities represent an important part of the genetic load. The aim of population cytogenetics is not only to estimate the frequency of cytogenetic abnormalities, but also their mutation rate, and the mechanisms that eliminate most of them and thus bring about a relative equilibrium.

The development of banding techniques has not only permitted a description or a better definition of some abnormalities but also the description of most of the nonpathological variants of the chromosomes (heteromorphisms). But, as pointed out by Vogel and Motulsky[58], the analysis of gene mutation at the molecular level and the development of banding techniques have blurred the distinction between chromosome and gene mutations. At the molecular level deletions, insertions, and duplication are possible.

1. The frequency of chromosome abnormalities

1.1. Abnormalities of chromosome number. These are by far the most frequent chromosome anomalies observed in human populations. Tables 1 and 2 show their frequencies in different populations.

Most of the numerical anomalies are caused by nondisjunction during meiotic divisions. Trisomies represent around 50% of the anomalies detected in abortuses and most of those detected in newborns.

In most trisomies and particularly in trisomy 21 a strong association is noted between the frequency of the disorders and maternal age. To explain this relationship many authors have made the assumption that the frequency of nondisjunction increases with age in the female meiosis. Recently it has been shown in trisomy 21 that maternal nondisjunction accounts for 80% of cases and paternal for 20%. The frequency of errors in the first meiotic division is 80% among the maternal cases and 60% among the paternal cases (pooled estimates are given by Juberg and Mowrey[33]). An elevated maternal age was noted in the four groups by many authors[33]. To explain these observations S. Aymé and A. Lippman-Hand[7] have recently formulated the hypothesis that there is decreasing maternal selection against affected conceptuses with advancing age.

Table 1. Frequencies of numerical chromosome anomalies

	Autosomal anomalies (%)	Sex chromosome anomalies (%)	Polyploidies (%)	Total chromosome abnormalities (numerical and structural, %)
In newborns surveys	0.14	0.21	0	0.62
In induced abortions (5–12 weeks gestational age)	4.2	0.7	1.2	6.6
In perinatal deaths	3.0	1.2	0.35	6–8
In first-trimester spontaneous abortions	25–35	10–15	8–14	50–60

From Boué et al.[10] and Hammerton[27].

Table 2. Rate of chromosome abnormalities in newborn babies*

Sex chromosomes	
Males	1:365
Females	1:661
Autosomes	
Trisomies	1:694
Balanced structural	1:517
Unbalanced structural	1:1675
Total abnormalities	1:161

* from Hammerton[27].

1.2. Chromosome structural rearrangements. Their incidence is given in table 3. At birth the incidence of balanced structural rearrangements is about 2‰[27]. Half of the rearrangements are reciprocal and half are Robertsonian translocations. Inversions are rarely found, even with banding studies.

For rearrangements, recent studies have shown that, firstly, the reproduction fitness of individuals with balanced chromosome rearrangements is lowered, but there are differences between the different types[29]. Secondly, in segregation studies of structural rearrangements, it has been observed that in the offspring of individuals carrying a Robertsonian translocation the frequency of balanced newborn exceed that of newborns with normal karyotype[10].

1.3. The mutation rates of the different rearrangements. We can note (table 3) differences between the mutation rates observed in newborn infants and in prenatal diagnosis even if we consider the balanced rearrangements which are not involved in natural selection. Boué et al.[9] think that the population involved in prenatal diagnosis is a selected one, having a poor reproductive history.

Jacobs[30] in the conclusion of her paper concerning mutation rates of chromosome rearrangements suggested that 'in attempts to monitor the population for the effects of mutagenic agents that are known or suspected of causing breakage, spontaneous abortuses are the most obvious and rewarding population to study'. However, this type of monitoring is difficult and very expensive. For this purpose it is easier to monitor rates through prenatal diagnosis. Monitoring in this case is a by-product, but it is necessary to define an unbiased sample in this population.

2. Chromosome mutations and population genetics

2.1. Frequency at conception and natural selection. Investigations of human abortions have provided much information about the frequency of chromosome anomalies, but the question remains as to how the incidence of such anomalies in spontaneous abortion relates to their incidence at the time of fertilization. Estimates of this frequency are based on extrapolation of prenatal wastage data. It is generally conceded that the true value is not likely to be less than 20%[15]. The frequency at birth of chromosome abnormalities is 0.62%. Among them most are biologically or socially sterile. The only exceptions are XYY and XXX trisomies and balanced rearrangements. Thus although there is a huge quantity of chromosome abnormalities, most of them numerical, there is a nearly perfect selection against these genotypes, and they can be classified as lethal dominant mutations in most cases.

Table 3. Frequency and mutation rate for all structural rearrangements

Population	Frequency ($\times 10^3$)	Mutation rate ($\times 10^3$)	Mutation rate for balanced rearrangements ($\times 10^3$)
Live births[a]	2.42[c]	0.26	0.15
Spontaneous abortions[a]	18.16	4.53	0.35
Prenatal diagnosis[b]	5.46	1.32	0.75

[a] From Jacobs[30]; [b] From Boué et al.[10]; [c] 2.53 in the data given by Hammerton[27].

2.2. Chromosomal mutations and evolution. The relationships between chromosome abnormalities and evolution must be discussed at two levels: the first concerns especially human reproduction, the second the evolutionary process.

Jacobs[28] has suggested that the high level of chromosomally abnormal conceptions may serve as a mechanism for the biological control of reproduction. This mechanism, although not the only one which reduces sibship size and increases the interval between births, has a selective advantage especially in human populations, where the offspring requires a great amount of parental care for a long period. This mechanism may be one means of ensuring that the optimum rather than the maximum number of offspring are born in a sibship. This attractive hypothesis cannot be studied in terms of population genetics, because we know too little about the genetic mechanisms, if indeed they exist, that regulate the rate of chromosomal abnormalities.

The role of chromosomal rearrangements in evolution was discussed by Dutrillaux[19]. In a recent review of the problem he considered different categories of rearrangements, depending on their supposed reduced fitness.

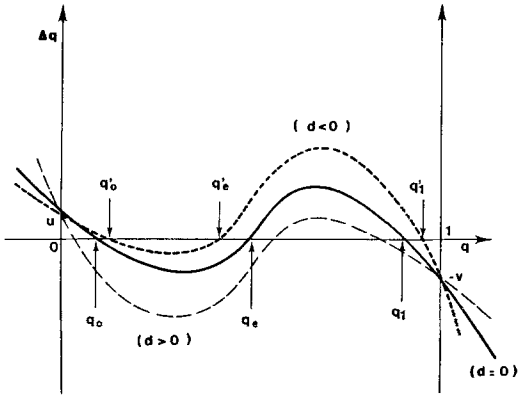
It is well known that nonfavored rearrangements such as pericentric inversions may be fixed only in small populations where stochastic processes like genetic drift occur. But one should not forget that a new neutral rearrangement will be eliminated in a large population, while a new favored rearrangement has only a slight probability of being fixed in a large population or even a small one.

According to Crow and Kimura[16], the probability of the fixation of a fresh mutation is given by

$$p = \frac{1 - e^{-2s}}{1 - e^{4Ns}}$$

where s is the selective advantage and N the effective size of the population ($P = 1 - e^{-2s}$ in large population).

A segregation distortion effect in favor of balanced gametes versus normal ones in individuals with a balanced rearrangement, cannot produce a large increase of the rearrangement frequency. As shown by Serre et al.[51] the segregation distortion is only followed by a shifting of the selection mutation equilibrium. So the fixation of such a rearrangement needs the occurrence of a stochastic process to draw the frequency of the rearrangement far from the equilibrium frequency q'_0 to an unstable equilibrium q'_e in order to reverse the sign of Δq (fig.). It must be noted that it was not possible to show that chromosomal rearrangements play a direct role in the modification of the phenotype by a position effect. But they can be one of



Shifting of the selection-mutation equilibrium when segregation distortion favors either normal gametes (d > 0) or balanced gametes (d < 0). In this last case the possible stable equilibrium frequency of the balanced rearrangement is q'_0. The distance between q'_0 and q'_e, the possible unstable equilibrium frequency is shorter than the distance q_0-q_e, (the stable and unstable disequilibrium when there is no distortion, (d = 0)). In conclusion, the greater the segregation distortion effect (d F 0), the lesser the required stochastic effect in order to pass beyond the unstable equilibrium frequency.
u is the mutation rate of the normal gamete to the balanced gamete; v is the reciprocal mutation rate. q is the frequency of the balanced gamete, Δq the change of this frequency per generation, due to selection effect. When the q'_e point overpassed, Δq > 0 so the rearrangement may be nearly fixed ($q'_1 \simeq 1$).

the reproductive isolating mechanisms that permit the process of speciation and genic differentiation.
Other chromosomal mutations have a role in evolution/polyploidy, mostly in plants, and gene duplication, which are two processes by which the amount of DNA can increase.

The malaria hypothesis concerning sickle-cell anemia: is a reappraisal necessary?

In 1949, Haldane[26] suggested that thalassemia could be maintained at a high frequency in some populations by a selective advantage of heterozygotes; the heterozygotes have an increased resistance to malaria caused by *Plasmodium falciparum* (Pf). This attractive hypothesis was adaptated by Allison[3] in 1954 to explain the high frequency of sickle-cell anemia in many regions of Africa. Allison 'found that in indigenous East Africans the sickle-cell trait affords a considerable degree of protection against subtertian malaria'. Evidence supporting this theory includes[13,43]:
- a differential survival of $\beta^A\beta^S$(AS) individuals. This observation is in favor of heterozygotes at advantage, but is not obligatorily related to malaria.
- a *Plasmodium falciparum* parasitemia lower in AS than in AA subjects.
- a near absence of severe malaria infection (cerebral malaria) in AS children.
- a geographic correlation between the distribution of malaria and the frequency of the sickle-cell gene, although the correlation is not constant.

However, since 1954, some contradictory results have been reported (Livingstone[39] and Le Coeur[38] for review). Carnevale et al.[11,12] have studied the relationships between the sickle-cell trait and Pf malaria in the Republic of the Congo. The study includes the General Hospital of Brazzaville and the populations of two villages (Djoumouna (D) and Linzolo (L)) which are located 20 kilometers southwest of Brazzaville. In this region there is stable and permanent malaria characterized by a high transmission rate contrasting with a relatively low plasmodic index. The frequency of the β^S globin gene is about 10–12%.
The main results of this study are as follows. The frequency of AS individuals does not change with age in Djoumouna; but in Linzolo a greater frequency of heterozygotes is noted among adults over 30 years old but not in younger adults. Plasmodic index was estimated in children under five years of age during longitudinal surveys in D. and L. These surveys included 89 AA and 20 AS individuals in D.,and 74 and 26 respectively in L. In D. the average plasmodic index is 30.6% in AA and 38.8% in AS individuals. In L. these figures are respectively 24.5% and 37.5%. These results do not show increased susceptibility to Pf infections among AA subjects. Parasite densities were studied in asymptomatic carriers during the longitudinal survey in D. and in young children hospitalized in the General Hospital. No difference between AA and AS individuals was observed.
Cerebral malaria was studied in the Department of Pediatrics of Brazzaville General Hospital. 2560 children were examined in 1977–1978; among them fewer than 1% had cerebral malaria. The frequency of AS carriers was 12.2% among 41 patients hospitalized in 1977, 1978 and 1980. This figure should be compared with the frequency of sicklers in the region (20–22%). These data are not sufficient to explain the maintenance of a high frequency of the S gene through a selective advantage of the AS subjects against malaria.
The results of these malaria surveys cannot explain the relatively high frequency of the Hb S gene. However, these data are in agreement with those reported by Ringelhann et al.[46], who noted that Hb S carriers had the highest infection rates. Moreover other authors have reported nonsignificant infection differences between AS and AA subjects in many African countries[38,39].
On the other hand these data can be explained by the hypothesis of Raper[45], who predicted in 1960 that intense transmission leads to a relaxation of natural selection against AA individuals. This hypothesis, however, cannot entirely account for the high frequency of the S gene observed in Central Africa.
All these contradictory results show that the relationship between *P. falciparum* malaria and the sickle-cell gene are not simple; a reappraisal of these relationships should thus be made, based on the specificity of each epidemiological situation; it is now well etablished that there is heterogeneity of malaria situations (stable/unstable, permanent/seasonal, etc), and also based on the genetic background of each studied population.
One of the causes of the contradictory results observed is perhaps α-thalassemia.
Recently, Lallemant et al.[36] have shown that α-thalassemia 1 and 2 are frequent in the region of Brazzaville. These authors have estimated the frequency of the (-α) gene as around 34%. If α-thalassemia confers some protection against malaria, it is difficult to demonstrate any increased resistance of AS individuals to malaria because more than 50% of the AA individuals have α-thalassemia 1 or 2. Moreover recent studies have shown that

α-thalassemia modifies the phenotype of sickle-cell anemia, diminishing the severity of the clinical and biological syndrome[22]. These findings need to be confirmed in African populations but are in favor of interactions between the β^S gene and α-thalassemia. It will be worthwhile to study simultaneously the relationships among malaria, the sickle-cell trait and α-thalassemia.

The DNA polymorphism

Since the introduction by Southern[53] of a method for DNA analysis, many DNA polymorphisms have been described. Southern's method includes restriction endonuclease digestion of the genomic DNA, electrophoretic separation of the DNA fragments and identification of specific DNA fragments by molecular hybridization. A single stranded radioactive nucleic acid is used to locate the genomic DNA sequences complementary to it. These strands are called probes. Probes may be made of genomic or complementary DNA. Synthetic oligonucleotides can be used as probes to detect a single nucleotide mutation[44]. Many genetic variations of restriction endonuclease sites have been described with this methodology. The addition or loss of the restriction sites results in different sized DNA fragments after digestion. These fragments are used to detect the genetic variations of restriction endonuclease sites. They are called restriction fragment length polymorphisms (RFLP). Sequencies data suggest that approximately one in every 100 or 200 nucleotides in the regions of DNA not involved in coding sequences is polymorphic. By extrapolation some 10^7 DNA polymorphisms may exist in man. Few of these polymorphisms are involved in a restriction site[32]. The RFLP behave like mendelian genes and can be used for linkage analysis or population genetics studies.

The study of sickle mutation (β^S) which in the homozygote state is responsible for sickle-cell anemia is an example of the use of DNA polymorphism in population genetics.

The β^S gene is frequent in Africa but it is also present in certain populations of Asia, Arabia and Southern Europe. Until 1978 the distribution of the gene was considered to be compatible with a single origin. In 1978, Kan and Dozy[34] observed that the β globin gene was associated with two different HpaI recognition sites; the β^S gene is associated either with a 7.6 kilobase (kb) fragment or a 13.0 kb fragment. The normal gene β^A is associated predominantly with a 7.6 kb fragment. A strong linkage disequilibrium between the β^S gene and the 13 kb RLFP can be noted. In 1980, Kan and Dozy[35] showed that the 13.0 and 7.6 kb fragments associated with the β^S gene have different geographical distributions. It was postulated by these authors that the mutation arose independently on chromosomes possessing either of the two RLFP. However, a recombination event can explain the existence of the two haplotypes[52].

In order to distinguish between the two hypotheses Wainscoat et al.[59] studied the polymorphic sites 5′ and 3′ to the β^S gene in 122 Jamaican patients with sickle-cell disease. 7 polymorphic sites were analyzed, 5 belonging to the 5′ haplotype group and 2 to the 3′ haplotype group. It has been shown in Jamaica that the β^S mutation is present on many different haplotypes but two of them are very frequent. Antonarakis et al.[4] and Wainscoat et al.[59] have found in normal β^A chromosomes that although the polymorphic sites within the 5′ and 3′ haplotypes are nonrandomly associated, the two groups of haplotypes are themselves in equilibrium. In contrast, Wainscoat[59] noted a strong linkage disequilibrium between 5′ and 3′ haplotypes of β^S chromosomes. For Wainscoat et al., all these findings are in favor of multiple origins for the βS mutation.

Recently, Antonarakis et al.[6] in a similar study found the β^S gene on 16 different haplotypes, which were defined by 11 restriction polymorphic sites. The three most common haplotypes are only rarely seen in the chromosomes bearing the β^A gene. Many hypotheses can explain the finding of the β^S gene in many different haplotypes[6]. One is that the $\beta^A \rightarrow \beta^S$ mutation occurred independently in each haplotype. Another hypothesis is that the β^S mutation occurred once and spread to the other haplotypes by means of repeated crossing-over events or interallelic undirectional gene conversion events. On the other hand, the real explanation may lie between the two alternatives.

Antonarakis et al.[5] have given evidence for multiple origins of another mutant globin gene, the β^E gene, in Southeast Asia.

Haplotype analysis associated with mutant genes in various populations should allow more precise analysis of the problem of the origin and spread of these mutations. DNA polymorphism is becoming a powerful tool in human population genetics.

The effect of relaxation of natural selection

1. The long term genetic effects of treatment of genetic diseases

This is a major preoccupation for many people. The basis of their concern can be illustrated by retinoblastoma which is a malignant tumor of the eyes, occurring as both inherited and noninherited types. Untreated, this disease leads to death, while surgery and radiotherapy lead to cure in most cases. In the next generation, this cure leads to an increase of the incidence of the inherited form. It can be shown, however, that the increase is slow, being determined by mutation pressure. G. Fraser[24] has shown that the dysgenic effect of medical treatment on the prevalence of a genetic disease can be studied very simply, whatever the mechanism of mendelian inheritance. If a disease is maintained by recurrent mutation, the prevalence of the disease will stabilize when a new equilibrium is reached at

$$x_1 = \frac{x_0(1-f_0)}{1-f_1}$$

where x_0 is the original prevalence and f_0 and f_1 are the relative fitness of individuals with the disease before and after treatment is instituted. Assuming that $f_0 = 0$ and $f_1 = 0.5$, then $x_1 = 2x_0$, and the prevalence is doubled. In the case of an autosomal recessive disease maintained by heterozygote advantage, the new equilibrium is reached at

$$x_1 = \left[\frac{1-f_0}{1-f_1}\right]^2 x_0$$

But we must note that the number of generations necessary to double the frequency of a deleterious autosomal

Table 4. Dysgenic and eugenic implications of prevention and of treatment, and of some technological and sociological changes affecting reproductive patterns, in the case of diseases determined by Mendelian inheritance or by chromosomal aberrations (D = dysgenic; E = eugenic)

	Autosomal dominant inheritance	Autosomal recessive inheritance	X-linked recessive inheritance	Chromosomal aberration
Treatment				
Increased fertility	D	D	D	D
Prevention				
Avoidance of marriage between heterozygotes by pre-marital screening		D		
Counselling leading to reproductive restraint	E	E	E	E
Ante-natal diagnosis and selective abortion of affected fetuses	E	D	D D-all males	D-translocations E-others
Others				
Reduction of heterozygote advantage		E	E	
Lowering of parental age at reproduction	E[a]			E[a]
Increase in mutation rate due to radiation and chemical agents	D	D	D	D

[a] Mutation rates are in some diseases age dependant. From Fraser modified[24].

gene responsible for a dominant or recessive disease and maintained by recurrent mutation is:

$$n = \frac{1}{\mu} \text{Log} \frac{1-q_0}{1-2q_0}, \ \mu \text{ is the mutation rate.}$$

When q_0 and μ have the same order of magnitude $n \simeq \frac{q_0}{\mu}$.

For a lethal recessive gene $\mu = q^2$ and $n = \frac{1}{q}$.

For example, if $q_0 = 1/100$, which is the frequency of the gene responsible for phenylketonuria in some populations, the treatment of all cases will double the gene frequency in 100 generations (about 2500 years).

A major increase of the disease prevalence will take only a few generations in the case of an autosomal dominant disease, but the rate of increase will be considerably slower in sex-linked recessive diseases and very slow in autosomal recessive ones.

2. The long term effects of prevention

This has been also studied by G. Fraser[24]. The effects are different when we consider genetic counselling, prenatal diagnosis and selective abortion of affected fetuses, and premarital screening of heterozygotes. Table 4 summarizes the dysgenic and eugenic effects of treatment and prevention.

In most cases the effects of prevention on the prevalence of the diseases is small. Retrospective genetic counselling followed or not by prenatal diagnosis has very little impact on the frequency of the diseases concerned. Only prospective procedures such as premarital screening of heterozygotes followed by avoidance of marriage or prenatal diagnosis reduce the incidence of diseases. We can note that in this case despite a dysgenic effect the incidence of the disease is notably reduced (theoretically the frequency drops to zero).

Genetic epidemiology

Genetic epidemiology is a branch of genetics whose goal is to study the genetic component of diseases. Its methods are those of population and formal genetics. Vogel and Motulsky[58] called it 'clinical population genetics' emphasizing the relationships among population genetics, clinical genetics and genetic epidemiology. Genetic epidemiology is mainly concerned with the study of familial clusters of disease. However, since a familial cluster may be due to a familial environment, it was necessary to introduce a new factor into the analysis, i.e., cultural inheritance[14]. The methods used in genetic epidemiology are numerous. We shall describe rapidly two of them: segregation analysis and the relationships between genetic polymorphism and diseases.

1. Segregation analysis

R. C. Elston[20] has defined segregation analysis as the statistical methodology used to determine from family data the mode of inheritance of a particular phenotype especially with a view to elucidating single gene effects. It is thus, as pointed by Elston[20], a basic tool in human genetics. Segregation analysis is based on a mathematical model that has four major components[20]:

– the joint genotypic distribution of mating individuals. For example under random mating the genotypic distribution for two alleles at an autosomal locus is given by the Hardy-Weinberg law.

– the relationship between phenotype and genotype. The phenotype is a qualitative or a quantitative character.

– the mode of inheritance: from one generation to the next it can be summarized by the genotypic distributions of the offspring conditional on the two parental genotypes.

– the sampling of the data.

With these components it is possible to devise a model for segregation analysis. Two major models are used in segregation analysis: a) *The mixed model* was first considered by Elston and Stewart[21] but was developed by Morton and MacLean[42]. The model associates a major locus with Mendelian transmission, a polygenic component and a random environmental effect. Inference of a major gene proceeds by rejecting the hypothesis of no major gene[37]. b) *The general transmission model* was proposed by Elston and Stewart[21]. Using this model, it is possible to test the agreement of transmission probabilities with Mendelian expectations. There is no polygenic component in this model. The hypothesis of no major gene coincides with that of no parent-offspring resemblance. Recently, Lalouel et al. have proposed a unified model[37]. Segregation analysis has seen many advances in the last decade both in the generality of the statistical models used and in the complexity of the family structure to which it is applied[49]. Although these methods have seen

limited application they have produced some interesting findings: a major locus was identified in the transmission of hypercholesterolemia and hypertriglyceridermia, and in the control of immunoglobulin E levels[49], and it has been shown that some forms of congenital glaucoma are a recessive disease[18].

2. Genetic polymorphism and diseases

The search for an association between genetic polymorphism and specific diseases is one of the methods used in studying the genetic component of disease. The first studies concerning the group ABO were made in the 20s. In 1953, the first association was described; it concerns phenotype A of the ABO blood group and cancer of the stomach[1]. In 1954, a stronger association was found between group 0 and peptic ulcer[2]. Since then many studies have been made concerning the ABO and Rh blood groups, the secretor and nonsecretor phenotypes, the Pi system and especially the HLA system. The diseases involved are numerous.

Population studies are generally the method used for studying whether a genetic polymorphism is associated with a disease. It consists of comparing the frequency of a genetic character, for instance an antigen of the ABO blood group or of the HLA system, in a group of patients and in a group of healthy controls. For each genetic character a 2×2 table is set up and a statistical test (usually a χ^2 test) is performed to detect an association (table 5).

Some statistical problems are raised by this type of study. In each study, many genetic characters are used for comparison and more than 20 HLA antigens are compared between the patient and control groups. Statistical analysis must take into account the number of comparisons analyzed. Combination of data is one of the ways to overcome the problem. Woolf[60] has described a method that permits analysis when several sources of data are available. Woolf's method also permits detection of heterogeneity between the sources.

This type of study involves several possibilities for drawing false conclusions. Biased ascertainment of patients or controls are the main source of error. It must also be recalled that an association between two characters does not imply linkage between the corresponding loci and that conversely, linkage between two loci does not imply association at the population level between the corresponding characters.

Many associations involving the ABO and HLA systems have been reported[25, 54, 58]. Those involving HLA antigens are 'stronger' than those involving ABO blood groups. 'Stronger' means a higher relative risk but this coefficient does not really measure the degree of association, since its level depends partly on the frequency of the characters studied. Many coefficients have been described to measure the associations but none is totally satisfactory[54, 55]. Nevertheless the data suggest that the contribution of HLA types to the system causing these diseases is greater than that of ABO groups to diseases found to be associated with them[58]. Many recent reviews[54, 58] list the different associations and the relative risk for each. For HLA the association includes ankylosing spondylitis, insulin-dependent diabetes mellitus, coeliac disease, psoriasis and multiple sclerosis, among others.

Table 5. Study of an association: the 2×2 table

	Character Present	Absent
Patients	a	b
Controls	c	d

The χ^2 test compares the frequencies of the character in patients $\left(\frac{a}{a+b}\right)$ and in controls $\left(\frac{c}{c+d}\right)$. The relative risk is the ratio $\frac{a \times d}{b \times c}$.

2.1. Genetic mechanisms involved in the associations with genetic polymorphism and disease. The most probable mechanisms involved in ABO blood group associations is that ABO genes form a part of a polygenic system which determines the genetic susceptibility to a disease. The small contribution of ABO genes reinforces the concept of multifactorial systems based on the effects of many genes[25].

For HLA the associations observed between HLA determinants and disease may be due to the direct effects of the identified associated determinants. For McDevitt and Bodmer[8, 41], the association is more likely to be due to the effects of alleles at closely linked loci which must be in significant linkage disequilibrium with the detected alleles at the HLA-A, B, C or D loci.

Family studies must complete the case control studies: classical linkage analysis if there is a hypothesis concerning the mode of inheritance of the disease; sib pair studies which analyze the HLA haplotype shared in sib pairs both of whom are affected[17, 56]. This latter method can give some insight into the mode of inheritance of disease.

It must be noted that none of the HLA associations with disease are absolute. One of the reasons for the lack of complete association is that a locus outside the HLA region is involved in the genetic susceptibility to the specific disease[8].

In the future the most interesting studies will concern the associations between HLA determinants and infectious diseases[57]. As pointed out by Bodmer[8] 'it seems likely that natural selection has molded the distribution of the HLA determinants in different populations but through associations with important infectious diseases rather that with the chronic diseases'. There are already hints in this direction, for example of a possible HLA association with leprosy[50]. Studies of newly discovered polymorphisms (DNA polymorphisms, new HLA markers) will lead to the discovery of new associations. Recently, many authors have reported associations concerning non-insulin-dependent diabetes and the risk of atherosclerosis and DNA sequences flanking the insulin gene[40, 47]. If confirmed, these associations would also show DNA polymorphism to be a powerful tool in genetic epidemiology.

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0014-4754/86/101102-08$1.50 + 0.20/0

The genetics of human reproduction

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Key words. Gonadal failure; impaired gametogenesis; human reproduction; meiosis; aneuploidy; maternal age effect.

Introduction

The attainment of full fertility in any organism requires the proper development and functioning of the reproductive system and survival of the foetus to term. Factors which operate to interfere with any of these processes will disturb the normal pattern and lead to partial or complete infertility.

The reproductive disorders of man are many and varied, but it is the contribution made by genetic anomalies which will be considered in this review. Mutations are known which affect gonadal development and sexual differentiation as well as those which act to disturb gametogenesis. Chromosomal abnormalities too can disturb gametogenesis; they also contribute significantly to human foetal wastage.

Several previous reviews on various aspects of the subject have been written and to these, the reader is also referred[24, 28, 31, 47, 54, 77, 123, 143]. A review of the genetic causes of sterility in the mouse, a much more widely investigated species, has been given by Searle[122].

1. Gonadal failure and impaired gametogenesis

1.1 The effect of the sex chromosomes

Considering that the X and Y chromosomes play a key role in sex determination and differentiation, it is hardly surprising that mutations at the gene loci involved, or sex chromosome imbalance, will produce serious consequences for the development of the reproductive system. Hermaphroditism, sex reversal and infertility are all well-documented features[54, 123].

In human cytogenetics, the discovery that sex chromosome aneuploids like 45, X and 47, XXY, could be associated with infertile syndromes, led to the initiation of chromosome surveys among selected groups of individuals experiencing reproductive problems. In one of the earliest studies, Jacobs et al.[79] examined the karyotypes of 32 women who had never menstruated spontaneously. Sex chromosome abnormalities were found in half of them: These included six with an XO genotype, five with sex chromosome mosaicism, three with a morphologically abnormal X chromosome, and two with an XY sex chromosome complement. This high level of sex chromosome abnormality compared with a frequency of only 1.8 per 1000 among newborn females[78]. The association of a male (XY) genotype with a female phenotype characterizes the two conditions of sex reversal, 'testicular feminization' and 'pure gonadal dysgenesis'[47]. The defect in XY females heterozygous for the X-linked Tfm (testicular feminization) gene[97] is well known, this mutation acting at the most fundamental level to disturb sex differentiation. In pure gonadal dysgenesis, gonadal failure is the primary defect and streak gonads are usually found[47]. In the XO female with Turner's syndrome and the XXY male with Klinefelter's syndrome, gonadal failure appears to relate to germ cell atresia. Recent insights into the role of the sex chromosomes in gamete survival in the mouse have been gained[93]. It would appear that shortly after birth in that species, a second X chromosome blocks male and facilitates female germ cell development. Oocytes at this time are known, both in man[52] and the mouse[42], to have two active X chromosomes, and germ cell loss in XO females may therefore relate to deficiency of X gene products[12, 89]. In XO human females, germ cell loss occurs principally around the time of birth[23, 124], the adult ovary being represented generally by a streak gonad. Nevertheless, limited fertility is achieved by some Turner individuals who retain oocytes over a much reduced reproductive span[83]. Germ cell loss in XY females with pure gonadal dysgenesis may also relate to deficiency of X-linked products.

The presence of two X chromosomes in germ cells of the mouse appears to be compatible with initiation of development in the male direction but is not compatible with spermatogenesis[93]. The XX component in XX/XY chimaeras appears to degenerate just before birth[94], while in XX, Sxr (sex reversed) males, degeneration sets in soon after birth[25]. On the other hand, spermatogenesis does take place in XO, Sxr males, some germ cells surviving to form spermatids and even a few abnormal spermatozoa[25]. In keeping with these findings, spermatogonia in much reduced numbers have been recorded in the testes of pre-pubertal XXY boys[98], but by puberty, little sign of spermatogenic activity remains. Gonadal failure resembling that seen in adult XXY men is also seen in XX