

Down's Syndrome and Maternal Inbreeding*

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The recent cytogenetic surveys (Hamerton *et al.*, 1965; Chitham and MacIver, 1965; Tonomura *et al.*, 1965; Kamada *et al.*, 1965) have clearly demonstrated that the overwhelming majority of cases with Down's syndrome both in Europe and Japan are of primary trisomy for the 21st autosome, a condition attributed to non-disjunction either at gametogenesis or at an early cleavage of the fertilized egg. Theoretically, this event may be caused by factors present either on paternal or maternal side, or on both sides, but the relative importance of the maternal side is generally accepted, since there is a strong dependency of the occurrence of this abnormality upon the maternal, not the paternal age if the two are appropriately separated (Penrose, 1933; Sigler *et al.*, 1965; Matsunaga, 1966). Therefore, it is necessary to search for the causal factors, both exogeneous and endogeneous, in the mothers who have born the affected children.

Of genetic interest is the question whether specific mutant genes can produce non-disjunction in the gonads of homozygous females. In *Drosophila melanogaster* a recessive gene was found to suppress crossing-over in oögenesis, resulting in the formation of diploid or aneuploid eggs (Gowen, 1933). In humans, this question could be answered by investigating consanguinity in parents of mothers of trisomics. If the supposed genes were not common, increased frequency of consanguinity would be expected among maternal than among paternal grandparents. Thus, Penrose (1961) found that, in a series of over 600 cases of Down's syndrome, ten of the mothers had cousin parents, while five of the fathers had such parents. Obviously the result was inconclusive but still suggestive for the hypothesis proposed. However, a recent report from Sweden (Forssman and Åkesson, 1966) based on more extensive data gave an apparently negative result.

Japan is well known for the high frequency of consanguineous marriages. Though the frequency varies according to the localities studied, it is usually about 2-5 per cent for urban areas and of the order of 10 per cent for rural areas. This situation may offer better opportunities to test the hypothesis of recessive genes for non-disjunction. The purpose of this paper is to describe the results of our investigation, based primarily on official records, and undertaken as part of epidemiologic studies of Down's syndrome in Japan.

* This paper is dedicated to Professor Dr. O. Frhr. von Verschuer in honour of his seventieth birthday. Contribution no. 618 from the National Institute of Genetics, Japan.

Material and methods

The index children with Down's syndrome were selected from the files of 834 cases that had been used in the previous study (Matsunaga, 1966). These cases, born in the years 1947-57, were ascertained in various institutions for mental defectives distributed all over Japan. Diagnosis had been made either by doctors associated with the institutions or by psychologists at the Children's Counselling Center prior to their entrance into the institutions, and only those cases with clear clinical signs of Down's syndrome were used.

Of the 834, 108 cases were drawn because they were born to mothers under 30

Generation

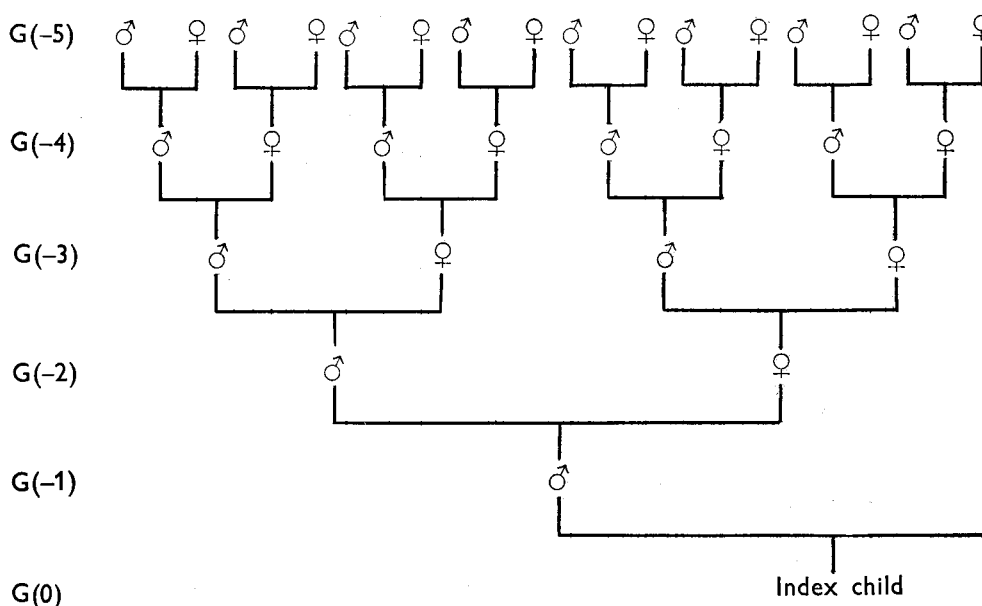


Fig. 1. Number of ancestors of the index child to be identified, in paternal line only

years old, and their permanent addresses were outside of Tokyo. The reason for this choice was that younger mothers of Down's children were more suspected to have a hereditary tendency to non-disjunction, and the prospect of obtaining sufficient informations on their ancestors seemed to be better than for older mothers; such prospect seemed also to be better in the ward offices outside of Tokyo, where the records were largely lost during the War.

Having ascertained the permanent address of the family, letters were sent to the

respective ward office asking for "koseki" records of their ancestors so far as the tracing was possible. These records were concerned with registration of family members, comprising their relationship and dates of birth, marriage and death. The task required for reconstructing the family trees is of course laborious, since the number of ancestors to be identified increases exponentially with ascending generations (Fig. 1). However, fairly sufficient informations could have been collected for 104 of the 108 cases, so that their data were analyzed in this paper. All these cases were apparently of sporadic occurrence in the families.

Results

Referring to Fig. 1, Tab. 1 represents the number of ancestors of the index children who could be identified. If one ancestor in G(-3) generation could not be identified, this would result in two such ancestors in G(-4) generation, and four in G(-5). The point is to see if the amount of information on which the identification of consanguinity is to be based differs significantly between the paternal and the maternal lines. This may be tested by comparing the rate of failure to identify

Tab. 1. Result of identifying ancestors of 104 children with Down's syndrome

Generation	Line	Total no. of possible ancestors	No. of ancestors identified	Rate of failure
G(-2)	Paternal	208	204	.019
	Maternal	208	204	.019
G(-3)	Paternal	416	402	.034
	Maternal	416	393	.055
G(-4)	Paternal	832	640	.231
	Maternal	832	637	.234
G(-5)	Paternal	1664	504	.697
	Maternal	1664	530	.681

the ancestors. In G(-3) generation, 14 ancestors could not be identified in the paternal line, while there were 23 such ancestors in the maternal line. Since the total number of possible ancestors in each line in G(-3) generation is $4 \times 104 = 416$, the rates of failure were respectively 0.034 and 0.055 in paternal and maternal lines, the difference being not significant ($\chi^2 = 2.29$, $P > 0.10$). In G(-4) and G(-5) generations, the rates were close to each other. Therefore, the comparison of consanguinity in grandparents between paternal and maternal lines may be justified.

The results of the frequencies of various types of consanguinity identified are summarized in Tab. 2. Among 104 index children, 6 had related parents, while 10 of

Tab. 2. Number and types of consanguinity identified in parents and grandparents of 104 children with Down's syndrome

Relationship	First cousins and their types				First cousins once removed	Second cousins	Total
	I	II	III	IV			
Parents	1	0	1	1	2	1	6
Grandparents							
Paternal line	3	1	1	4	0	1	10
Maternal line	4	1	2	1	2	2	12

the fathers and 12 of the mothers had such parents. The apparent lower frequency of consanguinity in the parents than in the grand-parents may be a reflection of the secular trend in this country. It is to be noted that there was virtually no difference in the frequency of consanguinity in the grandparents between paternal and maternal lines. When four types of first-cousin marriages were compared, their respective frequencies also showed no difference between the two lines.

Discussion

Before evaluating the above results, we have to examine what proportion of the mothers of Down's children should have consanguineous parents if the hypothesis of a recessive gene for non-disjunction holds true. Let the presumed gene be a with frequency q in the population, then the frequency of aa females from unrelated parents is $\frac{1}{2} q^2$, and the frequency of such females from consanguineous marriages is $\frac{1}{2} [Fq + (1-F) q^2]$, where F is the inbreeding coefficient for the offspring from these marriages. Specifically, the value of F is $\frac{1}{16}$ for children from first-cousin marriages, so that the above frequency is equal to $\frac{1}{2} \left(\frac{q}{16} + \frac{15}{16} q^2 \right)$. For the sake of simplicity, we assume that first-cousin marriages occur in a fraction c of all marriages while other remotely consanguineous unions are of negligible frequency.

Then, in a sample of aa females that were ascertained as mothers of Down's children, the expected proportion of those with cousin parents, k , is expressed by the following formula:

$$k = \frac{\frac{1}{2} c \left(\frac{q}{16} + \frac{15}{16} q \right)}{\frac{1}{2} (1 - c) q^2 + \frac{1}{2} c \left(\frac{q}{16} + \frac{15}{16} q^2 \right)} = \frac{c (1 + 15 q)}{c + 16 q - c q}$$

We may now estimate the k , first for Penrose's data previously mentioned and then for the Japanese data. The estimate of q can be obtained from the incidence of Down's syndrome at birth. This is well known for Europeans to be about 1 per 600 total births, so that q may be of the order of 0.04. On the other hand, the value of c may be replaced by the observed frequency of cousin marriages among paternal grandparents of Down's children. In Penrose's data this is about 0.01. Substituting the two estimates in the above formula, we obtain the estimate of k as 0.025. Thus, the expected number of mothers with cousin parents in Penrose's series of over 600 cases is about 15, which is not quite different from the 10 he actually found.

While in Japan the data on accurate incidence at birth of Down's syndrome are still insufficient, we consider, on rather indirect evidence, that it is probably close to that found in Europe and North America. However, since our cases were selected on account of younger mothers, we should use a lower value, say 0.025, for q . In our sample of 104 cases, 9 of the fathers had first-cousin parents, so that the value of c is about 0.09. Using the two estimates, k may be calculated as 0.254. This means that 26 mothers are expected to have first-cousin parents whereas only 8 such mothers were observed. The standard deviation of the latter value is 2.7, so that the great discrepancy between observation and expectation in our data appears to argue strongly against the hypothesis proposed.

In the above considerations, the mutant gene has been assumed to relate to a single autosomal locus. If there were different major genes on several loci, and the homozygosity for each of them could independently produce the same result of non-disjunction, then the individual gene frequency should become lower and hence the expected proportion of inbred mothers among those of Down's children would become higher. If sex-linked genes were involved, types I and II of first cousin marriages (cf. Tab. 2), where the coefficients of inbreeding of the offspring for sex-linked genes, F' , are $\frac{3}{16}$ and $\frac{1}{8}$, respectively, would occur more frequently in maternal than in paternal grandparents. In the absence of any such increase, our results indicate that neither autosomal nor sex-linked recessive genes are likely to have been responsible for non-disjunction, at least in the mothers who had born Down's children at a younger age.

Summary

In order to test the possible effect of maternal inbreeding upon non-disjunction, ancestors of 104 children with Down's syndrome born to younger mothers were identified on the basis of official records. Among them, 6 had related parents, while 10 of the fathers and 12 of the mothers had also such parents. The relationship comprises first-cousins, first-cousins once removed, and second cousins. It was concluded that recessive genes are unlikely to be involved in the aetiology of non-disjunction of the 21st autosome, at least in younger mothers.

Acknowledgement

This work was supported by grants from the Rockefeller Foundation (RF 61113) and the Toyo Rayon Foundation for the Promotion of Science and Technology.

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RIASSUNTO

Al fine di valutare il possibile effetto dell'endogamia materna sulla non-disgiunzione, sono stati identificati, sulla base dei registri ufficiali, gli ascendenti di 104 bambini affetti da Sindrome di Down, nati dalle madri più giovani. Sei di questi avevano genitori consanguinei, e così pure 10 padri e 12 madri. Il rapporto comprende cugini primi, cugini 1½ e cugini secondi. Si conclude che è improbabile che i geni recessivi siano responsabili dell'eziologia della non-disgiunzione del ventunesimo autosoma, per lo meno nelle madri più giovani.

RÉSUMÉ

Afin d'évaluer la possibilité d'un effet de l'endogamie maternelle sur la non-disjonction, l'on a identifié, sur la base des enregistrements officiels, les ancêtres de 104 enfants atteints de Syndrome de Down, nés aux mères les plus jeunes. Des parents consanguins furent trouvés pour six d'entre eux, ainsi que pour 10 pères et 12 mères. Le rapport comprenait premiers cousins, cousins 1½, deuxièmes cousins. En conclusion, il paraît improbable que les gènes récessifs soient responsables de l'étiologie de la non-disjonction du 21^{ème} autosome, tout au moins en ce qui concerne les mères les plus jeunes.

ZUSAMMENFASSUNG

Um die Hypothese zu prüfen, dass die Homozygotie der rezessiven Gene bei den Müttern eine ätiologische Rolle für « Non-disjunction » spielen könnte, wurden die Forfahren von 104 Patienten mit Down's Syndrom, die von jüngeren Müttern geboren waren, auf Grund offizieller Urkunde identifiziert. Unter den 104 Patienten hatten sechs blutsverwandte Eltern, während sich blutsverwandte Grosseltern 10mal auf der väterlichen und 12mal auf der mütterlichen Seite befanden. Bei der Beziehung der Verwandtschaften handelte es sich um Vetternehen 1., 1½. und 2. Grades. Der Verfasser kam zu dem Schluss, dass rezessive Gene für die Entstehung von « Non-disjunction » des 21. Autosoms wahrscheinlich keine Bedeutung hatten, wenigstens nicht bei jüngeren Müttern.