Endemic Goitre in Greece: A Study of 379 Twin Pairs*

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Non-toxic goitre is endemic in Greece (Malamos Koutras, Kostamis, Kralios, Rigopoulos, and Zerefos, 1966a), and detailed biochemical studies have implicated iodine deficiency as the main causative factor (Malamos, Miras, Koutras, Kostamis, Binopoulos, Mantzos, Levis, Rigopoulos, Zerefos, and Tassopoulos, 1966b). However, in the endemic regions studied, both goitrous and non-goitrous persons are iodine deficient practically to the same extent (Malamos *et al.*, 1966b), which agrees with results in other parts of the world (Wayne, Koutras, and Alexander, 1964), most notably in Western New Guinea (Choufoer, van Rhijn, Kassenaar, and Querido, 1963). This has prompted a search for additional factors which predispose a part of the population to goitre development, or to put it in another way, which protect the other from it.

It is now recognized that some cases of sporadic goitre, with or without cretinism, are due not to iodine deficiency but to inherited defects in the biosynthesis of thyroid hormones (McGirr, 1960; Stanbury, 1966; Wayne *et al.*, 1964). None of these defects has been proved to occur in endemic goitre (Murray and Stanbury, 1962), and a careful search in Greece has not produced evidence that any of these is of importance in the aetiology of the goitrous endemic (Malamos *et al.*, 1966b). Nevertheless, since the environmental factor of iodine deficiency does not explain entirely the development of endemic goitre, this has revived interest in the possible implication of genetic factors, which may potentiate the effects of a mild environmental iodine deficiency. Hadjidakis, Koutras, and Daikos (1964) have shown that in Greece endemic goitre affects some families more than others from the same village. This has been interpreted as

evidence of an inherited factor, though the influence of the small family environment cannot be excluded.

In a subsequent study by Malamos *et al.* (1966a) the familial aggregation of goitrous cases has been confirmed. However, it has been found that not only are the progeny of goitrous persons more extensively affected than the descendants of non-goitrous parents, but also that there is a statistically significant concordance between husbands and wives. Women married to goitrous men are themselves more often goitrous than the wives of the non-goitrous inhabitants of the same village. This, of course, points to the small family environment rather than to heredity as the cause of the familial aggregation of cases.

In view of these inconclusive findings, the present investigation was undertaken. 379 twin pairs from the endemic regions of central Greece were examined for the presence or absence of goitre, and the results were analysed separately for the monozygotic and the dizygotic twins, in order to find out whether there was a higher concordance in the former, as would be expected if goitre was an inheritable characteristic.

Subjects and Methods

Visits were made to 109 goitrous villages in three administrative districts (Table I). A sample of the population (usually children and adolescents aged 10-20 years) was examined for goitre, in order to know the over-all prevalence in these regions. All the twins

TABLE I
POPULATION INVESTIGATED

	Administrative District		
	Eurytania	Larissa	Jannina
Goitrous villages studied	23	46	40
Population	15,614	60,488	33,713
Number of persons examined	404	2452	2159
Percentage of persons with goitre	54.5	50.8	52.6
Twin pairs	51	172	156

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living in these villages were examined clinically for the presence or absence of goitre, by the criteria previously used by us (Malamos *et al.*, 1966a). If one of the twins was not living in the village, the pair was excluded from the study.

The twin pairs were classified as monozygotic or dizygotic, on the basis of the general appearance and the following blood tests, which were performed at the Haematology Laboratory of the 'Alexandra' Hospital (Dr. Ph. Fessas): (1) ABO, (2) CDE, and (3) M, N.

Results

The general characteristics of the population investigated are shown in Table I. Table II summarizes the results, the statistical analysis of which is shown in Table III.

It is obvious from Table II that the group of dizygotic twins contains a higher proportion of discordant pairs (i.e. the one twin goitrous and the other non-goitrous) than the monozygotics, which are to a greater extent concordant (i.e. either both members of the pair goitrous or both non-goitrous).

TABLE II
RESULTS OF TWIN PAIRS EXAMINATION

		Number	Percentage
Monozygotic males	Both goitrous	9	16.7
	Neither goitrous	39	72.2
	Discordant	6	11.1
Monozygotic females	Both goitrous	16	40.0
	Neither goitrous	20	50.0
	Discordant	4	10.0
Dizygotic males	Both goitrous	13	18.3
	Neither goitrous	42	59.2
	Discordant	16	22.5
Dizygotic females	Both goitrous	17	25.0
	Neither goitrous	30	44.1
	Discordant	21	30.9
Dizygotic male and female	Both goitrous	25	17.1
	Neither goitrous	72	49.4
	Male goitrous	4	2.7
	Female goitrous	45	30.8

TABLE III
STATISTICAL ANALYSIS

		Concordant		Discordant		
		No.	%	No.	%	
Males	Monozygotic	48	88.9	6	11.1	$\chi^2 = 2.75$
	Dizygotic	55	77.5	16	22.5	$p = N.S.$
Females	Monozygotic	36	90.0	4	10.0	$\chi^2 = 6.26$
	Dizygotic	47	69.1	21	30.9	$p < 0.02$
Totals	Monozygotic	84	89.4	10	10.6	$\chi^2 = 8.97$
	Dizygotic	102	73.4	37	26.6	$p < 0.005$

N.S. is not significant.

However, in Greece there is a sex difference in the prevalence of endemic goitre (Hadjidakis *et al.*, 1964; Malamos *et al.*, 1966a), and for this reason dizygotic pairs of different sex have been excluded from the statistical analysis shown in Table III. This compares the results obtained in monozygotic twins with those obtained with dizygotic twins concordant as to sex, i.e. either both members of the twin pair males or both females, pairs consisting of one male and one female twin being excluded. Even so there is a difference in the concordance rate between monozygotic and dizygotic twins. This difference is statistically significant for female twins ($\chi^2 = 6.26$, $p < 0.02$) or for both sexes taken together ($\chi^2 = 8.97$, $p < 0.005$). For male twins there is only a suggestive trend ($\chi^2 = 2.75$, $0.05 < p < 0.10$).

If the concordant twin pairs are subdivided into those with both members goitrous and those with neither, the results are similar (χ^2 with two degrees of freedom: for males 3.08, $0.20 < p < 0.30$; for females 6.85, $p < 0.05$; for both sexes 8.97, $p < 0.02$).

Discussion

It is clear from the results presented above that monozygotic twins have a higher rate of concordance for the presence of endemic goitre than dizygotic twins. Twins of the same sex, either mono- or dizygotic, are supposed to share the same small family environment, and so the increased concordance is attributed to the greater genetic similarity characterizing the monozygotic twins. In fact this problem may be more complicated, since monozygotic twins may have a more similar environment than dizygotic ones. Nevertheless, the environmental factors leading to goitre are mainly dietetic ones, and twins either mono- or dizygotic presumably share the same food.

The decision whether twins are identical (monozygotic) or not is often very difficult, requiring examination of the placenta and complete blood group studies (Clarke, 1964). However, in a study of a large number of twin pairs such as this, an occasional error in classification would not be expected to alter appreciably the over-all results, and for this reason the criteria for monozygosity adopted here (general appearance and a limited number of blood tests) appear adequate.

The results of this study do not conform to those previously obtained from Switzerland by Eugster (1934), who reported that monozygotic twins did not show a greater concordance with respect to endemic goitre than did dizygotic ones. The reason for this discrepancy may be that in Switzer-

land the environmental factor (iodine deficiency) has been stronger than it is now in Greece, thus masking the possible contribution made by heredity, or that in Switzerland the population is genetically more homogeneous for the characteristics predisposing to goitre. Furthermore, endemic goitre should not be viewed as a single entity. Although iodine deficiency is generally accepted as the decisive factor in most endemics (Wayne *et al.*, 1964), nevertheless the dietary iodine intake and the urinary excretion values range from a few $\mu\text{g.}$ per day, as in New Guinea (Choufoer *et al.*, 1963), to levels generally considered as adequate, as in Eastern Kentucky (London, Koutras, Pressman, and Vought, 1965). In this latter case factors other than iodine deficiency are presumably involved, but their nature remains still elusive.

Taste sensitivity to phenylthiourea (PTC) has not been studied in the present work, since in Greece there is no relation between PTC taste sensitivity and endemic goitre (Malamos *et al.*, 1966a). An association between the non-taster status and sporadic non-toxic goitre has been previously reported by some workers (Harris, Kalmus, and Trotter, 1949; Kitchin, Howel-Evans, Clarke, McConnell, and Sheppard, 1959) but denied by Fraser (1963). In endemic goitre in Israel an association has been reported by Brand (1963), but in other countries this has not been the case (Fraser, 1963; De Luca and Cramarossa, 1965; Malamos *et al.*, 1966a).

The evidence from the present study, as stated above, points to heredity as playing a role in the familial association of goitrous cases previously reported by several authors (Trotter, Cochrane, Benjamin, Miall, and Exley, 1962; Hadjidakis *et al.*, 1964; London *et al.*, 1965; Malamos *et al.*, 1966a). It would be difficult, however, to define more precisely the nature of the genetic factors involved and how they affect the predisposition to endemic goitre. A frequency distribution diagram of the estimated thyroid gland sizes in the endemic areas of Greece does not show two peaks, but only a continuous unimodal curve (Malamos *et al.*, 1966a), thus excluding any simple mode of inheritance. Environmental factors such as iodine deficiency are more important than heredity, since, in order to prove a statistical difference no less than 379 twin pairs have been required in the present study, whereas if heredity was a more important factor there would be a more striking difference between mono- and dizygotic twins, readily recognizable even in a much smaller population sample. So it may be concluded that, though heredity seems to be indeed implicated in the predisposition to endemic

goitre, its role is probably a minor one, in comparison with the potent environmental factors involved.

Summary

In 109 villages in the endemic goitre areas of Greece 379 twin pairs were examined for the presence or not of goitre. There was a higher concordance rate between the members of the monozygotic twin pairs than in the dizygotic ones. The difference was statistically significant even if dizygotic twins of different sex were excluded. It is concluded that heredity plays a role, probably a minor one, in the predisposition to endemic goitre.

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