

Acta Genet Med Gemellol 33: 251-258 (1984) © 1984 by The Mendel Institute, Rome

TWIN RESEARCH 4 - Part B: Twin Psychology and Behavior Genetics Proceedings of the Fourth International Congress on Twin Studies (London 1983)

Finnish Twins Reared Apart II: Validation of Zygosity, Environmental Dissimilarity and Weight and Height

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Abstract. Within the Finnish Twin Cohort of like-sexed adult twin pairs, a subgroup of pairs separated at an early age has been identified. In 165 pairs, both cotwins responded to questionnaires in 1975 and 1979. An environmental dissimilarity score was formed which consists of items on whether the twins had lived after separation in the same community, attended the same school, were on the same grade at school, how often the cotwins met, how often they met common friends and relatives and whether they attended the same clubs etc, or not. To validate the zygosity diagnosis obtained by questionnaire in 1975, those pairs whose zygosity was unknown as well as those with the least contact after separation were contacted for blood sampling (11 bloodgroups). Of 15 pairs with no zygosity diagnosis, 10 responded (1 no address, 2 abroad, 2 refused). Six pairs were classified MZ and 4 DZ. In 12 MZ and 8 DZ pairs undergoing bloodgroup determination, the classification of only one pair changed from DZ to MZ. The following intraclass correlations for height and weight were found:

Age at	No. (Cases	Wei	ght	Hei	ight
separation	MZ	DZ	MZ	DZ	MZ	DZ
0 - 5 0 - 10	18 30	61 95	0.88 0.87	0.31 0.36	0.88 0.92	0.70 0.70

Key words: Twins, Rearing environment, Adoption, Behavior Genetics, Weight, Height.

INTRODUCTION

The aim of the study design of identical twins reared apart is to control for the genetic variance of a trait, so that only the family and environmental conditions vary. The remaining differences in MZ pairs can thus be attributable to the effects of environment.

The similarity of the MZ probands can be estimated using intraclass correlations which are commonly used for indication of broad sense heritability [4].

Earlier studies of twins reared apart have consisted of fairly small series of twin pairs: Newman et al [13] studied 19 pairs separated under the age of 7, Shields [16] 44 pairs separated under the age of 9, and Juel-Nielsen [7] 12 pairs separated under the age of 6. In addition, Bouchard [3] recently described a series of identical twins separated at a very early age [2,6]. A recent review of the reared-apart twin literature has been done by Farber [5].

The purpose of this project is to analyse the effect of environmental and genetic factors of health behaviour, personality and physical development among Finnish twins reared apart in childhood before the age of eleven years. In an earlier paper, the preliminary characterization of the rearing environment and causes of separation was given [12]. In this paper, zygosity determination, analysis of environmental similarity of cotwin, and genetical analysis of height and weight will be presented.

THE FINNISH TWIN COHORT STUDY

The sample was selected from the Finnish Twin Cohort Study. The latter consists of all Finnish adult same-sexed twin pairs (N = 17,357 pairs) born before 1958 and with both members alive in 1967. All pairs of persons with the same date of birth, same sex, same surname at birth and same community of birth were selected from the Central Population Registry of Finland. This selection procedure yielded the base population for the Finnish Twin Cohort Study [8]. Twinship was confirmed by a questionnaire study in 1975 and inquiries to local parish birth registers. The 1975 questionnaire covered health-related items and standardized measures of morbidity, with a total of 97 questions. The basic distributions of the questionnaire study have been documented [9,10,11]. The questionnaire was mailed to all pairs with both members alive in 1975 (N = 16,269 pairs). The overall response rate was 89%.

In addition to the questions on health related items, the twins were asked whether the twin partners are living together or not, and if not, at what age separation had occurred.

DEFINITION OF PRESENT STUDY GROUPS

The sampling procedures for the twins reared apart have been described in detail elsewhere [12]. This selection procedure yielded 478 Finnish speaking pairs. All those alive in 1979 and with adequate adress data were then sent, during November 1979 - January 1980, a questionnaire on their childhood environment. Some 30 Swedish-speaking pairs were excluded from the study. A total of 165 pairs replied to both questionnaires (1975 and 1979). The final study group (125 pairs, 30 MZA and 95 DZA) for this analysis was formed using a cut off point of an environmental dissimilarity scale (described below). Control groups were formed of twin pairs which have been reared together at least up to 16 years of age and not living together in 1975 (MZT, DZT). The mean age at separation among the study groups was as follows: MZA 4.3 yr, DZA 4.2 yr, MZT 23.0 yr and DZT 22.5 yr. The final study groups and their mean ages by sex, zygosity and separation status are presented in Table 1.

Mean age at			Men		Women			Total		
	separation	Mean	SD	N	Mean	SD	N	Mean	SD	N
MZA	4.3	38.8	13.6	13	48.8	13.7	17	44.5	14.3	30
DZA	4.2	43.3	13.5	35	44.9	13.8	60	44.3	13.7	95
MZT	23.0	40.8	13.9	23	52.9	17.1	24	47.0	16.6	47
DZT	22.5	41.9	14.0	63	44.1	16.7	72	43.1	15.5	135

TABLE 1 - Mean Age (yr, 1975) by Sex, Zygosity and Separation Status

TABLE 2 - Blood Test Results for Twins Reared Apart

		Questionnaire diagnosis				
		MZ	DZ	XZ*		
Blood test diagnosis	MZ DZ	12 0	1 7	6 4		
Nonparticipants		5	7	2		
Total		17	15	12		

* Three pairs were not requested being abroad or address being unavailable.

TABLE 3 - Blood Test Participation	in Relation to	Zygosity,	Age-at-Separation	and Environmental
Similarity Score				

Environmental	Blood	Age	0-5 yr	Age 6	-10 yr
similarity score	test	MZ	DZ	MZ	DZ
	No	4	14	5	8
< 16	Yes	1	1	2	0
≥ 16	No	4	51	10	34
<i>≥</i> 16	Yes	14	10	2	0

TABLE 4 - Item Analysis of Environmental Dissimilarity Score

Item	Correlation between item and sum score
Different communities	0.56
Different schools	0.65
Different classes	0.63
Frequency of cotwin contact	-0.84
Frequency of contact of common friends	-0.86
Frequency of contact of same relatives	-0.77
Participation in same clubs, etc.	-0.44

Reliability coefficient = 0.82 Range of scores 7-10

ZYGOSITY DETERMINATION

In the whole Finnish twin cohort, zygosity was determined by a highly accurate questionnaire method, that has been validated by blood markers [14,15]. Because the zygosity questions used mainly information on similarity in appearance and confusion by other people in childhood, the validity of this zygosity determination method in the sample of twins reared apart had to be evaluated separately.

All but 15 of the 165 pairs who replied to both questionnaires could be classified as MZ or DZ according to their responses to the 1975 questionnaire. The remaining 15 pairs were requested to deliver a blood sample to the Finnish Red Cross Blood Service. Three pairs were abroad or had no address and in two pairs there was nonparticipation of at least one cotwin. Eleven bloodgroups were determined from each sample: ABO (A1,A2,0), MNSs, Rh (CCwTEce), Fy (Fya,Fyb), Jk (Jka,Jkb), Kell (KkKpa), LW (Lwa, Lwb), Pl, Le/Se (Lea, Leb). The probability of misclassification for a twin pair drawn at random from the Finnish population for these bloodgroups was 0.078.

In addition, a sample of pairs classified as MZ or DZ by the questionnaire method was drawn to see if the questionnaire diagnosis could be used for the whole study group or whether bloodgroup determinations had to be done for all. The pairs selected had been separated mostly at the age of 0-5 years and had the least contact after separation. The blood samples were obtained as above. The result is shown in Table 2. One DZ pair was reclassified as MZ, the questionnaire diagnosis of zygosity being upheld in the other 19 pairs.

Table 3 shows the distribution of pairs according to their participation in the blood testing in relation to age at separation, environmental similarity score and zygosity. Of the MZ pairs selected into the final analyses in this study only 4 out of 18 separated at ages 0-5 years were not blood sampled.

ANALYSIS OF ENVIRONMENTAL SIMILARITY OF COTWINS

The final study group was formed using an environmental dissimilarity scale of seven items from the questionnaire on family environment and childhood. These were items on whether the twins had lived after separation in the same community, attended the same school, or been on the same grade at school, and on how often the cotwins met, how often they met common friends and relatives, and whether they attended the same clubs, etc, or not. The responses were recorded so that a high score indicated a greater dissimilarity of environmental contact after separation. The summation was carried out for each cotwin's variables separately. The reliability of the environmental dissimilarity scale was 0.83 in a sample of 383 pairs of twins separated prior to age 17. The correlation of individual items to the sum score was high. The highest correlation coefficients were for frequency of contact between cotwins (-0.84) and contact between common friends (-0.86). The lowest correlation was for participation in the same clubs, organizations, etc (-0.44) (Table 4).

For studying if age, sex, zygosity, age at separation and causes of separation have an effect on environmental similarity, the correlation of the environmental similarity score to these variables was examined (Table 5).

When background variables were considered, zygosity had a slight negative correlation in twins separated under 11 years of age indicating that MZ twins had minimally greater environmental dissimilarity than DZ twins. A slight positive correlation to sex was found,

Variable	Corre	lation
variable	Cotwin 1	Cotwin 2
Sex	0.066	0.054
Age	-0.049	0.003
Zygosity	-0.075	0.094
Causes of separation		
Maternal death	-0.008	-0.032
Paternal death	-0.015	-0.031
Maternal illness	0.075	-0.145
Paternal illness	-0.044	-0.098
Divorce	-0.064	-0.065
Single parenthood of mother	0.027	0.000
Cotwin illness	-0.063	-0.139
Own illness	-0.101	-0.125
Economic conditions	-0.003	0.187
Reason unknown	-0.029	-0.112
Cotwin score	0.770	0.756

 TABLE 5 - Correlation Analysis of the Environmental Dissimilarity Score by Sex, Age, Zygosity and Causes of Separation. Pairs separated prior to age 11 (N=165) and both respondent.

TABLE 6 - Mean Weight (kg) by Sex, Zygosity and Separation Status Among Individual Twins (First member of pair)

	M	en	Women		
	Mean	SD	Mean	SD	
MZA	75.0	12.0	60.9	8.8	
DZA	74.7	10.2	63.6	10.2	
MZT	71.7	7.3	59.0	7.6	
DZT	74.7	10.3	61.2	9.8	

TABLE 7 - Mean Height (cm) by Sex, Zygosity and Separation Status Among Individual Twins (First member of pair)

	Me	en	Women		
	Mean	SD	Mean	SD	
MZA	174.5	6.5	157.2	6.8	
DZA	173.4	7.0	160.0	5.5	
MZT	171.4	5.8	160.4	4.2	
DZT	172.8	6.2	160.6	5.3	

that is, greater environmental dissimilarity in female twins. The age correlation was close to zero.

The correlations to causes fo separation were also very low. The highest correlations were the causes of either twin's illness or parent's illness, but not death (Table 5).

ANALYSIS OF WEIGHT AND HEIGHT

Weight and height are derived from the 1975 health questionnaire and are thus based on self-report at that period. They were recorded to the nearest kilogram and centimeter, respectively.

To assess whether the twins reared apart from an early age were as individuals comparable with respect to study variables to twins reared together, the mean values of individuals were analysed by sex and study group. The mean weight (Table 6) varies among men between 71.7 and 75 kg and among women between 59 and 63.6 kg. There was no statistically significant difference between groups. The mean height (Table 7) varies among men between 171.4 and 174.5 cm and among women between 157.2 and 160.6 cm. There was no statistically significant difference between groups.

For weight, mean intrapair differences, correlations and heritability estimates are shown in Table 8. Among the MZA pairs, men have slightly smaller intrapair differences, higher correlation and higher heritability estimate than women. MZA men show a stronger correlation (0.89) than MZT men (0.79), but among women the effect is reverse (0.71 vs 0.77).

Also for height men have slightly smaller intrapair differences, stronger correlations and a higher heritability estimate than women (Table 9). MZA pairs show a slightly smaller correlation than MZT pairs.

DISCUSSION

This sample of twins reared apart is derived from a representative nationwide twin cohort. The data is derived from two questionnaire studies, and thus the analysis is based only on the respondents to both studies. The study series was then restricted to those in which intrapair contact after separation was minimized. This was done to obtain as "pure" a sample of separated pairs as possible. Zygosity was determined using the questionnaire method and control blood testing that this is very sensitive and seems to work even when there has been only little contact after separation.

The environmental dissimilarity was measured using a 7-item scale. The reliability of the scale was high. The causes of separation had a slight influence on the environmental dissimilarity. The highest correlations were the causes of either twin's illness or parent's illness. Thus, when illness in the family was reported, environmental dissimilarity was greater. This is understandable, because the environmental similarity score was formed mainly from variables of social contacts between twin partners.

Both weight and height show high heritability estimates, slightly higher for men than for women. The estimates of common environmental effect did not differ significantly from zero. Family studies in the USA [1] and Finland [17] have found much lower heritability estimates for weight and height. These results are similar to those obtained in earlier studies on twins reared apart [5] or reared together.

	Mean intrapair difference (kg)		Corr	elation	N of pairs	
	Men	Women	Men	Women	Men	Women
MZA	4.62	4.94	0.888	0.712	13	16
DZA	8.42	10.1	0.338	0.169	33	54
MZT	4.35	3.92	0.792	0.768	23	24
DZT	9.51	8.67	0.281	0.330	61	70
Veighted		h ² =	0.87	0.68		
east		$SE(h^2) =$	0.06	0.10		
quares		$c^{2} =$	-0.11	0.05		
		$SE(c^2) =$	0.08	0.11		
		$\chi^2 =$	0.70	2.26		
		P =	0.71	0.32		

Table 8 - Intrapair Differences and Correlations for Weight

Table 9 - Intrapair Differences and Correlations for Height

	Mean intrapair difference (cm)		Correlation		N of pairs	
	Men	Women	Men	Women	Men	Women
MZA	1.62	2.13	0.934	0.872	13	15
DZA	5.03	5.07	0.541	0.333	33	54
MZT	1.65	1.38	0.941	0.941	23	24
DZT	4.63	4.45	0.502	0.425	60	71
Weighted		$h^2 =$	0.935	0.874		
east		$SE(h_1^2) =$	0.035	0.057		
squares		$c^{2} =$	0.007	0.063		
		$SE(c^2) =$	0.041	0.060		
		$\chi^2 =$	0.439	1.36		
		P =	0.80	0.51		

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Aknowledgment. This study has been supported in part by a grant from The Council for Tobacco Research USA-Inc., and by a grant from the Yrjö Jahnsson Foundation.

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