Acta Genet Med Gemellol 33:375-381 (1984)

TWIN RESEARCH 4 - Part C: Clinical Studies
Proceedings of the Fourth International Congress on Twin Studies (London 1983)

Environmental Effects on Cardiovascular Risk Factors in Chinese Adolescent Monozygotic Twins

C.J. Chen[1], B.H. Cohen[2], E.L. Diamond[2], T.M. Lin[1], J.S. Chen[3]
[1] *Institute of Public Health, National Taiwan University College of Medicine, Taipei, Taiwan;* [2] *Department of Epidemiology, The Johns Hopkins University School of Hygiene and Public Health, Baltimore, Maryland; and* [3] *Department of Clinical Pathology, National Taiwan University Hospital, Taipei, Taiwan*

Abstract. The monozygotic (MZ) cotwin control method was employed to elucidate possible environmental determinants of systolic blood pressure (SBP), diastolic blood pressure (DBP), serum cholesterol and triglyceride levels. A population-based twin sample of 73 male and 77 females MZ twin pairs was recruited from 12 junior high schools in Taipei city. Intrapair differences in blood pressure were negatively associated with intrapair difference in vegetable preference, attaining significance for DBP in males and SBP in females. Cholesterol was positively associated with milk consumption and preference for sweets, fried foods, meat and fish. A negative association was also observed between choleserol and vegetable preference. These associations for cholesterol were significant in males only. Triglyceride level negatively associated with preferences for sweets and vegetable, attaining significance for vegetables in both males and females and for sweets in males only.

Key works: Blood pressure, Cholesterol, Triglycerides, Diet, Cotwin Control Method

INTRODUCTION

Twin studies can provide valuable information on the relative and interactive contribution of genetic and environmental components in cardiovascular disease (CVD). The conventional comparison between monozygotic (MZ) and dizygotic (DZ) twins, followed by estimation of genetic variance and heritability, is based on the assumption that environmental correlations are equal for MZ and DZ twins. This assumption has been questioned, among others by Lilienfeld [15], and Smith [22], from his study of the relative intrapair similarity of MZ and same-sexed DZ twins on selected socioenvironmental factors, concluded that the role of environment needs to be more fully evaluated. After critically reviewing the assumptions of classical twin studies, Dibble et al [9] and Elston and Boklage [10] suggested that, besides the estimation of genetic variance and heritability, twin studies should have an entirely different and more promising use.

The MZ cotwin control method and the twin environmental method provide opportunities for additional insight into disease: any difference in an attribute between MZ cotwins should be the result of intrapair differences in environmental factors. This method is similar to a natural experiment, consisting in the examination of a possible relationships between environment and the attribute studied, with genetic influences controlled.

The purpose of this report is to present the results of a MZ cotwin control study of the dietary effects on major CVD risk factors: systolic blood pressure (SBP), diastolic blood pressure (DBP), serum cholesterol and triglyceride levels in Chinese adolescents.

MATERIALS AND METHODS

Data collection

Twin sampling, zygosity determination, and data collection procedures have been extensively described elsewhere in this issue [3]. Shortly, blood pressure and serum cholesterol and triglyceride levels were examined in a population-based sample of 73 male and 77 female MZ twin paris. (DZ pairs were also studied but are not considered in the present note). A life style questionnaire and a Chinese version of the Junior Eysenck Personality Inventory [11] were also administered and information on family background and early life experience of the twin subjects was obtained from the parents.

Methods of Analysis

Multiple regression analysis was employed to explore the possible effects of environmental factors on CVD risk factors of MZ twins. The model of the multiple regression analysis of intrapair difference is as follows:

$$D_i = \beta_1 d_{i1} + \beta_2 d_{i2} + \ldots + \beta_j d_{ij} + \ldots + \beta_k d_{ik} + e_i$$
$$\text{for } i = 1, 2, \ldots, n$$
$$\text{and } j = 1, 2, \ldots, k$$

where D_i is the intrapair difference in dependent variable (ie, CVD risk factor) of the i^{th} MZ pair; d_{ij} is the intrapair difference in the j^{th} independent variable (ie, host or environmental factor) of the i^{th} MZ pair; and e_i is the error term of the i^{th} MZ pair.

Two members of a given pair were randomized into groups I and II, the intrapair difference in each variable was derived from abstracting the value of cotwin in group II from that of cotwin in group I. The difference might thus be positive, zero or negative. These intrapair differences were used in the multiple regression analyses to keep the directionality of the association. If a regression coefficient is negative, it implies that the relationship between dependent variable and that specific independent variable is inverse. For example, if most twins A have higher cholesterol levels and lower vegetable preference than twins B (ie, most intrapair differences in cholesterol levels are positive and most differences in preference to vegetables are negative), there will be a negative regression coefficient for vegetable preference. There are two indications of a negative regression coefficient: (1) the greater the absolute intrapair difference in independent variable, the greater the absolute difference in dependent variable; and (2) the intrapair difference in independent variable is negative while the difference in dependent variable is positive, and vice versa.

As both twins of a given pair have identical demographic variables, family history of CVD, familial relative food frequency and maternal use of low salt diet during twin pregnancy, these variables were not included in regression equations. Cigarette smoking was also excluded because only a few twin pairs smoked cigarettes.

In order to adjust for possible effects of intrapair differences in anthropometric characteristics, personality inventory, serum chemistry, and activity levels on the intrapair differences in SBP, DBP, cholesterol and triglyceride levels, these adjustment variables were also included in the multiple regression equation. Details of these methods are described elsewhere in this issue [3].

RESULTS

Blood Pressure Levels

Both multiple correlation coefficients and standardized regression coefficients of intrapair differences in dietary preferences and beverage consumption included in the multiple regression analyses of SBP and DBP are shown in Table 1. After including all independent variables in the multiple regression equations, the multiple correlation coefficients were 0.70 for SBP and 0.66 for DBP in males, and 0.75 for SBP and 0.82 for DBP in females.

Significant positive associations were found between SBP and meat preference, and between DBP and animal organ preference in females but not in males. However, the direction of these associations between blood pressure levels and preferences for meat and animal organs was inconsistent in males vs females and in SBP vs DBP. Intrapair differences in DBP and SBP were negatively associated with vegetable preference in both males and females, but these relationships attained statistical significance only for DBP in males and SBP in females.

The intrapair difference in DBP was positively associated with soft drink consumption for females but not form males. In males only, a negative association between SBP and coffee consumption, and a positive association with alcoholic beverage consumption were observed. However, the direction of the association between blood pressure and various beverage consumption was inconsistent in males vs females and in SBP vs DBP.

Serum Cholesterol Level

Table 2 shows multiple correlation coefficients and standardized regression coefficients of intrapair differences in dietary preferences and beverage consumption in the multiple regression analyses of serum cholesterol level. The analysis showed correlation coefficients of 0.94 for males and 0.60 for females after all the independent variables were included.

Intrapair differences in cholesterol levels did not show significant associations with intrapair differences in any of the food preferences for females. However, among males, significant positive associations were found between cholesterol levels and preferences for sweets, fried foods, meat and fish. While intrapair differences in cholesterol levels and vegetable preferences were negatively associated in both males and females, again statistical significance was attained in males only.

Intrapair differences in cholesterol levels were significantly associated with intrapair differences in all types of beverage consumption for males: milk, tea, coffee and alcoholic beverage consumption showing positive associations and soft drinks a negative one. In females, the only statistically significant relationship was inconsistent with the findings in males: a negative association between intrapair differences in cholesterol level and tea consumption. The only consistency in the direction of these associations for beverages was the one for milk consumption.

Table 1 - Correlation and Regression Coefficients for Intrapair Differences in Dietary Preferences in Regression Analyses of Blood Pressure in MZ Twins

	Male pairs (N = 73)		Female pairs (N = 77)	
	SBP	DBP	SBP	DBP
Multiple correlations coefficients[a]	0.70	0.66	0.75	0.82
Standardized regression coefficients				
Dietary preference				
Sweet foods	– 0.18	0.06	– 0.21	– 0.18
Salty foods	– 0.13	– 0.24	– 0.02	0.15
Fried foods	– 0.06	– 0.02	– 0.09	0.09
Meat	0.08	– 0.21	0.47**	– 0.09
Fish	– 0.23	0.17	– 0.12	– 0.11
Animal organs	0.24	– 0.02	– 0.00	0.57**
Eggs	– 0.20	– 0.09	– 0.21	0.19
Vegetables	– 0.15	– 0.31*	– 0.47**	– 0.15
Beverage consumption				
Milk	– 0.27	0.23	– 0.17	0.23
Soft drinks	0.30	– 0.20	0.15	0.50*
Tea	0.01	0.03	0.00	– 0.05
Coffee	– 0.37**	0.09	– 0.08	– 0.02
Alcoholic beverage	– 0.41**	0.09	– 0.15	0.17

a Intrapair differences in anthropometric characteristics, personality inventory, serum uric acid, magnesium and calcium levels, and activity levels were also included in the equation to adjust for their possible confounding effects.

* $P < 0.05$; ** $P < 0.01$

Table 2 - Correlation and Regression Coefficients for Intrapair Differences in Dietary Preferences in Regression Analyses of Serum Cholesterol in MZ Twins

	Male pairs (N = 73)	Female pairs (N = 77)
Multiple correlations coefficients[a]	0.94	0.60
Standardized regression coefficients		
Dietary preference		
Sweet foods	0.41**	0.09
Salty foods	– 0.04	– 0.04
Fried foods	0.32**	0.14
Meat	0.32**	0.16
Fish	0.20**	0.17
Animal organs	– 0.01	– 0.08
Eggs	– 0.09	– 0.15
Vegetables	– 0.26**	– 0.18
Beverage consumption		
Milk	0.67**	0.22
Soft drinks	– 0.26**	– 0.02
Tea	0.25**	– 0.38*
Coffee	0.48**	– 0.08
Alcoholic beverage	0.34**	– 0.11

a See note to Table 1.

* $P < 0.05$; ** $P < 0.01$

Serum Triglycerides Level

After all independent variables were included in the stepwise multiple regression alayses of intrapair difference in triglyceride level, the correlation coefficients were 0.84 for males and 0.61 for females (Table 3).

Triglyceride levels showed negative associations with vegetable prefences for both males and females and with sweets preferences for males only. Intrapair differences in triglyceride levels and fried food preference were positively associated in males. No significant relationship with intrapair differences in preferences for salty foods, meat animal organs or eggs were observed among either males or females.

Triglyceride levels were negatively associated with milk consumption in females only. In males, positive associations were found between intrapair differences in triglyceride levels and both soft drinks and tea consumption, while there was a significantly negative relationship with alcoholic beverage consumption.

Table 3 - Correlation and Regression Coefficients for Intrapair Differences in Dietary Preferences in Regression Analyses of Serum Triglycerides in MZ Twins

	Male pairs (N = 73)	Female pairs (N = 77)
Multiple correlations coefficients[a]	0.84	0.61
Standardized regression coefficients		
Dietary preference		
Sweet foods	– 0.22**	– 0.05
Salty foods	– 0.09	0.14
Fried foods	0.27*	– 0.15
Meat	0.14	0.03
Fish	– 0.07	0.16
Animal organs	0.16	– 0.24
Eggs	– 0.11	– 0.12
Vegetables	– 0.24*	– 0.40*
Beverage consumption		
Milk	0.03	– 0.55**
Soft drinks	0.32*	0.07
Tea	0.24*	– 0.02
Coffee	0.04	– 0.17
Alcoholic beverage	– 0.25*	– 0.00

a See note to Table 1.

* $P < 0.05$; ** $P < 0.01$

DISCUSSION

The fact that adolescent twins of a given pair in this study were living together implies that they were exposed to the same dietary background in their family. The major contribution to their intrapair difference in dietary exposure was thus the difference in their preferences.

In the analysis of the relationship between intrapair differences in blood pressure levels and environmental factors in MZ twins, other relevant, but more complex factors (such as anthropometric characteristics, personality, serum chemistry profiles, activity levels) were also included in the multiple regression equation to adjust for their effects.

Blood pressure levels were consistently negatively associated with vegetable preference; and the associations were significant for DBP in males and SBP in females. This finding is in agreement with a report by Sacks et al [21] who found that the mean blood pressure of 210 men and women eating mainly vegetable food was lower than that usually found in Western populations. Further study on this relationship may result in a better understanding of the causation of elevated blood pressure.

The association of blood pressure with increased sodium (salt) intake and urinary sodium excretion, long reported in experimental and epidemiological studies [1,4,6-8,12, 14,16-20], was not observed in this study. One possible explanation is that the intrapair difference in salty food preference in MZ twins was too small to cause a significant intrapair difference in blood pressure. The threshold concept of the response of blood pressure to sodium intake might be another explanation, ie, the effects of sodium are significant only when its intake exceeds a certain threshold level. The interaction between genetic and environmental factors might also be another possibility.

Several dietary preferences and beverage consumption pattern were found to be consistently associated with cholesterol levels. Only in males, however, did the associations attain statistical significance, a finding which may derive from the much greater similarity in these factors in female than male cotwins. That is, environmental factors were so similar in female MZ twins that any difference in their effects would be difficult to detect. There may also be other variables which influence serum cholesterol level in adolescent females and are not included in this study, such as hormonal and other factors associated with the onset of puberty and the menstrual cycle.

Intrapair differences in serum cholesterol were positively associated with the intrapair difference in milk consumption and preferences for sweets, fried food, meat and fish, while the association between serum cholesterol and vegetable preference was negative. These findings agree with several other studies which have shown associations of serum cholesterol with dietary cholesterol and saturated fats [2,5,13].

Intrapair differences in serum triglyceride levels were consistently associated with intrapair differences in sweets and vegetable preferences, as well as soft drink consumption, but not all in the same direction. Associations were negative with sweets and vegetable preferences, and positive with soft drink consumption. Very few other studies have focused on the dietary effects on triglycerides [2,5], and in those studies a positive association was found with carbohydrate content (ie, sucrose).

Acknowledgements: The authors wish to thank Drs. T.H. Beaty and C.A. Newill for their helpful comments and suggestions. This work was partly supported by NSC-70-0412-B002-41.

REFERENCES

1. Abboud FM (1974): Effects of sodium, angiotensin, and steroids on vascular reactivity in man. Federation Proceedings 33:143.
2. Berenson GS (1980): Cardiovascular Risk Factors in Children. New York: Oxford University Press.
3. Chen CJ, Cohen BH, Diamon EL, et al (1984): Genetic variance and heritability of cardiovascular risk factors in Chinese adolescent twins. Acta Genet Med Gemellol 33:363-373.

4. Coleman TG, Granger HJ, Guyton AC (1971): Whole body circulatory autoregulation and hypertension. Circulation Research 28/29: I-176.
5. Connor WE (1980): Cross-cultural studies of diet and plasma lipids and lipoproteins. In Lauer RM, Shekelle RB (eds): Childhood Prevention of Atherosclerosis and Hypertension. New York: Raven Press, p 99.
6. Crysant SG, Walsh GM, Kem DC, et al (1979): Hemodynamic and metabolic evidence of salt sensitivity in spontaneously hypertensive rats. Kidnoy International 15:33.
7. Dahl LK (1960): Possible role of salt intake in the development of essential hypertension. In Bock KD, Cottier PJ (eds): Essential Hypertension. 2nd International Symposium. Berlin: Springer-Verlag.
8. Dahl LK, Knudsen KD, Heine M, et al (1967): Effects of chronic excess salt ingestion. Genetic influence on the development of salt hypertension in parabiotic rats: evidence for a humor factor. J Experiment Med 126:687.
9. Dibble E, Cohen DJ, Grawe JM (1978): Methodological issues in twin study: the assumption of environmental equivalence. In Nance WE (ed): Twin Research: Psychology and Methodology. New York: Alan R Liss Inc., p 245.
10. Elston RC, Boklage CE (1978): An examination of fundamental assumptions of the twin method. In Nance WE (ed): Twin Research: Psychology and Methodology. New York: Alan R Liss Inc., p 189.
11. Eysenck SBG (1965): Manual of the Junior Eysenck Personality Inventory. London, University of London Press.
12. Freis ED (1976): Salt, volume and the prevention of hypertension. Circulation 53:589.
13. Keys A (1970): Coronary heart disease in seven countries. Circulation 41 (suppl 1): 1.
14. Kirkendall WM, Connor WE, Abboud F, et al (1976): The effect of dietary sodium chloride on blood pressure, body fluids, electrolytes, renal function, and serum lipids of normotensive man. J Lab Clin Med 87:418.
15. Lilienfeld AM (1961): Problems and areas in genetic-epidemiologic field studies. Ann New York Acad Sci 91:797-805.
16. Luft FC, Rankin LI, Bloch R, et al (1979): Cardiovascular and humoral responses to extremes of sodium intake in normal black and white men. Circulation 60:697.
17. Meneely GR, Batterbee HD (1976): High sodium-low potassium environment and hypertension. Am J Cardiol 38:768.
18. Oliver WJ, Cohen EL, Neel JV (1975): Blood pressure, sodium intake, and sodium related hormones in the Yamamo Indians: a "no-salt" culture. Circulation 52:146.
19. Page LB, Damon A, Moellering RC Jr (1974): Antecedents of cardiovascular disease in six Solomon Island societies. Circulation 49:1132.
20. Prior IAM, Grimley-Evans JG, Harvey HPB, et al (1968): Sodium intake and blood pressure in two polynesian populations. N Engl J Med 279:515.
21. Sacks FM, Rosner B, Kass EH (1974): Blood pressure in vegetarians. Am J Epidemiol 100:390.
22. Smith RT (1965): A comparison of socioenvironmental factors in monozygotic and dizygotic twins: Testing an assumption. In Vandenberg SG (ed): Methods and Goals in Human Behavior Genetics. New York, Academic Press, p 45.

Correspondence: Dr. Chien-Jen Chen, Institute of Public Health. National Taiwan University College of Medicine, Taipei, Taiwan 100.