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Using MZ Twins in Experimental Research to Test for the Presence of a Genotype-Environment Interaction Effect

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Abstract. Despite some evidence that genotype-environment interaction (G×E) effects may be involved in the variation observed in behavioral and biological traits, few attempts have been made to detect and quantify this component of genetic variation in humans. We propose that one way to achieve this goal is to challenge several genotypes in a similar manner, submitting both members of several MZ twin pairs to an ethically acceptable experimental treatment capable of inducing an adaptative response. In this situation, the G×E effect can be assessed with a two-way analysis of variance for repeated measures on one factor, the treatment effect. In this design, twins are considered nested within the pair, whereas the treatment effect is considered a fixed variable. The intrapair resemblance in the response to the treatment is quantified with an intraclass correlation coefficient computed with between-sibhips and within-sibhips means of squares. To illustrate this approach, changes induced by long-term endurance training were studied in 10 MZ twin pairs. Significant intrapair resemblance in the response of maximal oxygen uptake was observed, with about 7 to 8 times more variance between pairs than within pairs. This design with MZ twins may be helpful in the study of human variation for multifactorial phenotypes.

Key words: Twin models, Genotype-environment interaction, Repeated measures, Treatment effect, Endurance training

INTRODUCTION

In the study of multifactorial quantitative phenotypes, it is of interest to establish the relative importance of the additive genetic component and to test for the presence of a major gene effect, a maternal or a paternal influence, a sex-limited

effect, and other phenomena as well. It is also useful to know whether there is a genotype-environment interaction effect contributing to the phenotypic variance and, if possible, the extent of that effect.

Genotype-environment interaction refers to a situation in which the sensitivity of the individual to the environment or to given lifestyle factors depends on his or her genotype [3,8]. This effect is an interaction above the main linear effect of the genotype and of the environment-lifestyle components resulting from individual differences in the response to existing environmental and lifestyle conditions or to changes in such conditions.

Even though ways of testing for the presence of a genotype-environment interaction effect have been proposed in human genetic epidemiology studies [1,3-6], most of the models used in the field assume that the effect is equal to zero. This is of course not totally satisfactory, particularly in studies dealing with physiological or metabolic phenotypes where the genotype-environment interaction phenomenon seems to be ubiquitous [10,11].

These observations suggest that there is a need for a procedure that could be used to test for the presence of individual differences in the response to given environmental-lifestyle conditions and for the possibility that such differences are associated with a genotype-environment interaction effect. One strategy, albeit an imperfect one, is to use MZ twins experimentally exposed to an altered but identical environmental-lifestyle set of conditions for a prolonged period of time, ie, exposed to a standardized treatment. This design could yield important information on the interaction issue for physiological and metabolic genetic questions.

THE METHOD

Essentially, the procedure requires that both members of several MZ pairs be subjected to exactly the same experimental treatment. Assuming that there are individual differences in response to the treatment, within-pairs and between-pairs variances can be obtained.

Some of the conditions that are important for the outcome of that kind of study include the following: a) determine twin zygosity as precisely as possible; b) keep age variation at a minimum; c) use preferably twin pairs of only one gender or control for gender difference if male and female twin pairs are involved and depending upon the phenotype of interest; d) apply the treatment in exactly the same manner to all twins under rigorously controlled experimental conditions; and e) select phenotypes that are not, or are very little, affected by prior exposure to the environmental-lifestyle variable altered in the study. A variant of the method would be to use both MZ and DZ twins. However, the value of the DZ twins in providing a control over the common environment effect as in the classical twin study design is greatly diminished in the present method as all the twin individuals are exposed to similar environmental conditions for the duration of the experiment.

To a large extent, the procedure is similar to that used in animal genetics when testing for a genotype-environment interaction effect by comparing various

strains of a given species exposed to a given treatment or a set of experimental conditions. The main difference is that by using MZ twins, we have only 2 subjects per genotype. However, the number of genotypes (MZ pairs) can be relatively large depending on the demands of the research question.

The data can be analyzed by a two-way analysis of variance for repeated measures on one factor (the treatment effect).

	B_1	B_2
A_1	twin a_1 twin b_1	twin a_1 twin b_1
A_2	twin a_2 twin b_2	twin a_2 twin b_2
.		
.		
.		
A_p	twin a_p twin b_p	twin a_p twin b_p

Factor A is considered a random effect and stands for the genotype effect with p levels in which p is the number of pairs. Factor B is fixed and represents the treatment effect with q levels in which q would be equal to 2 if only pre- and post-treatment measurements are considered. Subjects are considered a third factor with two levels (2 subjects in each twin pair) and nested under factor A.

An Illustration: The Response to Exercise-Training

Several experiments have been performed in our laboratory to establish the importance of the individual differences among sedentary subjects in the response to a defined exercise-training stimulus [1]. In one study, designed to estimate the effects of endurance training on maximal oxygen uptake (VO_2 max), 13 women and 11 men were subjected to a fully standardized and laboratory supervised 20-week cycle ergometer training program [7]. These sedentary males and females improved their VO_2 max from 2.3 to 2.9 liters per min (l/min), with a mean gain of 0.6 l/min. However, when each individual's improvement in VO_2 max was computed, a standard deviation of 15% was associated with a mean training change of 30% and the variation in the training response ranged between 5% and 88%. The maximal aerobic power data reported per kg of body weight revealed the same variation in response to exercise-training. Therefore, there are considerable individual differences in the response of VO_2 max to exercise-training, some individuals exhibiting a high responder pattern, while others are almost non-responders. What is (are) the factor(s) responsible for such human variations in adaptation to exercise-training?

Briefly, age and gender of subjects, as well as their prior training experience, do not contribute much to human variation in relative trainability, at least in the range

from puberty to about 70 years of age. The major causes of human variation in the response to training appear to be the current phenotype level, ie, the pretraining status of the trait considered, and perhaps a genetically determined capacity to adapt to exercise training. The latter would represent the so-called role of heredity in trainability, ie, the genotype-training interaction.

To elucidate this phenomenon, several experiments were undertaken. In one study, 10 pairs of MZ twins were subjected to a 20-week standardized endurance training program [9]. In response to this program, mean VO_2 max of the subjects improved by 16%. As expected, there were considerable interindividual differences in response, as the training gains ranged from about zero to 40% for VO_2 max. Differences in the response to training were not, however, distributed randomly among the MZ twin pairs (Table). Thus, the intraclass correlation computed with the amount of training gain in VO_2 max (l/min) was 0.77, indicating that members of the same MZ twin pair yielded a fairly similar response to training. There was about 7 to 8 times more variance between genotypes than within genotypes for the response to training in terms of gains in liters of O_2. These results suggest that the sensitivity of maximal aerobic power to endurance training is largely familial and most likely genetically determined.

Table - Effects of exercise-training and intrapair resemblance in the maximal oxygen uptake response in 10 pairs of MZ twins[a]

Variable	Pre-training mean (SD)	Post-training mean (SD)	Training effect F ratio	Interaction effect F ratio	Intrapair resemblance response (intraclass)
VO_2 max (l O_2)	2.57 (0.70)	2.94 (0.78)	25.7**	7.8*	0.77*
VO_2 max (ml O_2/kg.min)	44.20 (6.00)	49.70 (5.90)	24.4**	6.8*	0.74*

[a] Adapted from Bouchard [1] and Prud'homme et al [9]. Intrapair resemblance in response after adjusting the data for the pretraining level became 0.75 for VO_2 max and 0.72 for VO_2 max per kg weight.
*$P < 0.003$; **$P < 0.001$

CONCLUSION

The method briefly described in this paper may be helpful in our efforts to understand the various determinants of the individuality in response to changes in lifestyle or environmental conditions. This approach can of course be used only with treatments that are ethically acceptable to volunteer MZ twins. The time commitment required of the twins is generally considerable but will obviously vary depending on the protocol of the study. We believe that the procedure can be

helpful in understanding the individual differences in the response to nutritional challenges, regular exercise, altitude exposure, as well as to other but carefully standardized and sustained conditions.

The model can also be used in conjunction with genetic markers at candidate genes for one or several phenotypes of interest. It is becoming increasingly clear that studies of candidate genes involved in multifactorial phenotypes [12] will contribute much toward the understanding of the genotype-environment interaction phenomenon and its biological significance. The present design, with its emphasis on the phenotypic response to an experimental and sustained challenge, along with the information available on the within-genotypes and between-genotypes variances, combined with allelic variation at candidate genes, may be helpful in our efforts to understand the biological basis of the individuality in the adaptation to changing environment and lifestyle.

REFERENCES

1. Bouchard C (1986): Genetics of aerobic power and capacity. In Malina RM, Bouchard C (eds): Sport and Human Genetics. Champaign, Ill: Human Kinetics, pp 59-88.
2. Berg B (1988): Variability gene effect on cholesterol at the Kidd blood group locus. Clin Genet 33:102-107.
3. Eaves LJ (1976): Human behavioural genetics. Proc R Soc Med 69:184-189.
4. Eaves LJ (1984): The resolution of genotype x environment interaction in segregation analysis of nuclear families. Genet Epidemiol 1:215-228.
5. Khoury MJ, Adams MJ, Dana Flanders W (1988): An epidemiologic approach to ecogenetics. Am J Hum Genet 42:89-95.
6. Lathrop GM, Lalouel JM, Jacquard A (1984): Path analysis of family resemblance and gene-environment interaction. Biometrics 40:611-625.
7. Lortie G, Simoneau JA, Hamel P, Boulay MR, Landry F, Bouchard C (1984): Responses of maximal aerobic power and capacity to aerobic training. Int J Sports Med 5:232-236.
8. Plomin R, DeFries JC, Loehlin JL (1977): Genotype-environment interaction and correlation in the analysis of human behavior. Psychol Bull 84:309-322.
9. Prud'homme D, Bouchard C, Leblanc C, Landry F, Fontaine E (1984): Sensitivity of maximal aerobic power to training is genotype dependent. Med Sci Sports Exerc 16:489-493.
10. Scriver CR (1988): Nutrient-gene interactions: The gene is not the disease and vice versa. Am J Clin Nutr 48:1505-1509.
11. Simopoulos AP (1988): Genetics and nutrition: Introduction to the symposium on human genetic variation and nutrition. Am J Clin Nutr 48:1497-1499.
12. Sing CF, Boerwinkle E, Moll PP, Templeton AR (1988) : Characterization of genes affecting quantitative traits in humans. In Wheir BS, Eisen EJ, Goodman MM, Namkoong G (eds): Proceedings of the Second International Conference on Quantitative Genetics. Sunderland, Mass: Sinauer Associates, pp 250-269.

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