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SHORT NOTE

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Possible Linkage Relationship Between Genetic Markers and Blood Magnesium and Zinc

A Twin Study

P. Darlu[1,2], E. Defrise-Gussenhoven[4], Y. Michotte[3], C. Susanne[4], J.G. Henrotte[2]

[1] Groupe de Recherches de Génétique Epidémiologique, INSERM, Paris; [2] R.C.P. Magnésium et Oligoéléments, CNRS, Faculté de Pharmacie, Paris; [3] Pharmaceutical Institute, and [4] Laboratorium Antropogenetika, Vrije Universiteit, Brussels

Abstract. In a sample of 63 like-sex dizygotic twins, red blood cell and plasma magnesium and red blood cell zinc concentrations were analyzed for linkage to each of 23 genetic systems by estimating correlation between proportion of genes identical by descent and biological resemblance for the trait. The results suggest possible linkage of red blood cell magnesium with the HLA locus and of red blood cell zinc with the GLO1 locus. However, studies applying more powerful tests are needed to confirm such conclusions.

Key words: Linkage, Magnesium, Zinc, Twins, Sib-pair method, Marker loci

INTRODUCTION

The quantitative inheritance of plasma and red blood cell magnesium (PMg and RBCMg) and red blood cell zinc (RBCZn) concentrations in man has previously been investigated [4-7]. The assumption that a major gene strongly influences RBCMg was supported by segregation analysis [18] and was also suspected for RBCZn [8], Association of HLA with RBCMg and also possibly with RBCZn have been suggested by Henrotte [15,16].

These results indicate that investigation of relationships between these quantitative traits and various genetic marker loci might be useful. As a first step, in this short note, we present results provided by applying the sib-pair method proposed by Haseman and Elston [12] for detecting linkage between a marker locus and a putative major gene.

SAMPLE AND METHODS

A sample of 63 liked-sex dizygotic twin pairs (33 males and 30 females) was collected in the course of

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a Belgian study organized by E. Defrise-Gussenhoven and Ch. Susanne from the Vrije Universiteit Brussels. The diagnosis of zygosity was performed on 24 systems by the late J. Brocteur and L. Kamenev at the Laboratoire des Groupes Sanguins, Liège. Details of this sample and on these systems have been published elsewhere [3,9,10]. Magnesium and Zinc concentrations, both in the plasma and red blood cell, were determined by atomic absorption spectrophotometry following a procedure previously described [6].

The sib pair method assumes that the difference between a sib pair in the metric trait is inversely related to the proportion of genes identical by descent (ibd) at the marker locus, under the assumption that the trait is explained mostly by a major gene near the marker locus: "The more similar the genotypes of sibs at the marker locus, the less will be the difference between them in the metric trait" [12].

The estimation of the proportion of genes ibd, p_j, for each blood group locus and for each twin pair, was calculated by the method of Haseman and Elston [12], using gene frequencies provided by the Centre de Transfusion, Liège (Director: A. André) from this area of Belgium; the HLA frequencies are from West Germany [13]. The proportion p_j for the jth sib pair is: $p_j = (f_{j2} + f_{j1}/2)$, f_{ji} being the probability that the jth pair has i genes ibd at the marker locus, given the sib-pair genotype.

The quantitative trait X is first adjusted for the well-known effect of age and sex [14] by regression on a fixed effect for sex and on a quadratic function of age. Then, the absolute difference d_j between the adjusted trait of the two dizygotic twins of the jth pair is calculated.

Finally, the Spearman rank correlation r_s is computed between p_j and d_j for each pair of the three traits for the 23 informative autosomal loci.

RESULTS AND DISCUSSION

The Table summarized the results. Out of 69 correlations tested, 4 are significantly negative and different from zero ($P < 0.05$, one-tail test). A lack of relation between the sibling similarity for the trait and the proportion of genes identical by descent should provide about a similar number. However, even if one linkage existed for a given trait, we could expect only one negative correlation more than those expected at random (say, 4 or 5 instead of 3 or 4 ...). Because we have no mean to distinguish between random correlations and correlation resulting from a real linkage, all the highest negative correlations can be discussed.

The sib-pair method is known to have low power in detecting an association [20,1,2]. Indeed, the power of this test increases with the sample size and is also dependent on the degree of linkage and on the genetic variance at the major locus. This variance is generally estimated from segregation analysis and is often found to be lower under a mixed model which takes into account polygenic inheritance [17,19] than under other models [11], as in the case of RBCMg [18]. Thus, the suspected power of a study is clearly related to the model used for segregation analysis. Even though the conditions for high power may be found the trait and marker loci could be distant enough so that a high negative correlation between p_i and d_i might not appear. Besides, we cannot estimate the recombination fraction by the method discussed because of the dependence between genetic variance and the genetic distance between trait and marker loci expressed in terms of recombination.

The efficiency of the method is also related to the number and the frequencies of alleles of the genetic marker. For the case where there is a dominance effect at the marker locus and where the frequency of the recessive allele is small, the power to detect linkage is substantially reduced [2]. So, the possible linkage between RBCZn and Inv ($r_s = -0.30$) must be cautiously assumed. For the same reason, we do not study the relationship with the Tf and G6PD systems for which all sib pairs have the same genotypes (except for one at

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G6PD and two for Tf). The relationship between PMg and Lewis is suspicious, especially since PMg is the variable with the lowest heritability and no major gene is suspected in its inheritance [7,18]. On the other hand, GLO1, ABO, and HLA-A, for example, provide "a priori" more powerful situations in which linkage can be investigated by this method.

The correlations between the RBCMg and both HLA-A and HLA-B were weak or not significant, but quite similar, and are the most negative correlations for RBCMg. They are consistent, since the two markers are linked and associations between some HLA-A and HLA B haplotypes and RBCMg have been reported elsewhere [15,16]. However, the negative correlation between RBCMg and Bf is not as high as expected with respect of the tightly link between HLA and Bf. Linkage between GLOl and RBCZn must also be taken into account, although a negative correlation between RBCZn and HLA, close to GLOl, should be also expected. Further investigation might explain this result. As discussed elsewere [18,8], such associations involving RBCMg and RBCZn concentrations have to be understood as a linkage between marker locus and a gene, which could be the marker locus itself. This gene may yield allelic forms of a protein or an enzyme (as cell-surface antigens for HLA and Glyoxalase I for GLO) acting differentially on permeability of the membrane of red blood cells to Mg and Zn or modifying characteristics of Mg or Zn binding proteins. This is the biochemical problem emphasized by these genetical observations.

Table. Spearman Rank Correlations between Proportion of Genes Identical by Descent (Pibd) and Intrapair Differences in DZ Twins for Plasma Magnesium d(PMg), Red Blood Cell Magnesium d (RBC Mg) and Red Blood Cell Zinc d (RBCZn) Concentrations According to 23 Autosomal Genetic Systems.

Genetic system	d (PMg)	d (RBCMg)	d (RBCZn)
ABO	0.044	0.204	0.068
RH	0.070	− 0.148	− 0.031
MNS	0.162	0.007	0.076
P	− 0.082	0.191	0.076
Le	− 0.322**	0.063	0.134
Fy	− 0.065	− 0.056	− 0.122
Jk	− 0.132	0.045	− 0.058
Hp	− 0.133	0.195	0.092
Gm	0.115	− 0.087	− 0.182
Inv.	− 0.012	0.171	− 0.300**
C3	− 0.108	0.323	0.233
Bf	− 0.027	− 0.092	− 0.006
ACPl	0.051	0.078	0.077
PGMl	0.016	0.242	− 0.121
AK	0.006	0.018	− 0.083
ADA	− 0.020	− 0.015	0.065
GPT	0.043	− 0.026	0.155
EsD	− 0.114	0.142	0.000
GLO1	− 0.046	0.164	− 0.294**
HLA A	− 0.141	− 0.223*	0.090
HLA B	0.016	− 0.186	0.110

Sample size = 63.
* $P < 0.05$; ** $P < 0.02$; one-tail t test.
Correlations between intrapair differences are not significant.

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Correspondence: Dr. P. Darlu, Groupe de Recherches de Génétique Epidémiologique, INSERM U.155, Château de Longchamp, 75016 Paris, France.

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