

ANALYSIS OF MIXTURE IN THE DISTRIBUTIONS OF IMMUNOGLOBULIN LEVELS IN A CHAGASIC POPULATION

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ABSTRACT

In a sample of three hundred individuals living in a Brazilian area where Chagas' disease is endemic, commingling in the distributions of IgA, IgG and IgM levels was investigated. Evidence of a mixture of two skewed components was found for IgG and IgM distributions due to a single gene effect or a discrete environmental event. One component was suggested for IgA distribution, which implies that both genetic and environmental determinants are multifactorial. The proportions of the upper components in the distributions of IgG and IgM levels were 0.31 and 0.12, respectively.

INTRODUCTION

Even though till now it has not been possible to demonstrate a major gene responsible for the regulation of immunoglobulin levels, there is evidence that genetic factors play an important role in the concentration of these proteins in human blood (McGue *et al.*, 1989). A single X-linked dominant gene has been hypothesized to explain the higher levels of IgM found in the females than in males (Grundbacher, 1972); a relationship between the number of X chromosomes and the level of IgM has also been detected (Rhodes *et al.*, 1969); and moderate values of heritability for both IgA and IgG have been estimated (Barbosa *et al.*, 1981a), suggesting a polygenic mode of transmission.

However, recent analyses of the distribution of immunoglobulin levels (McGue *et al.*, 1989) showed no evidence for commingling in the distribution of IgM and so, do

not support the major gene hypothesis; on the other hand the distributions of both IgA and IgG showed significant commingling, which may be due to the action of a major gene and/or a discrete environmental factor.

We made analyses of commingling of distributions of IgA, IgG and IgM levels in a Brazilian data set collected for genetic studies on Chagas' disease (Barbosa *et al.*, 1981b; Krieger *et al.*, 1983).

MATERIAL AND METHODS

Serum samples were collected from individuals living in a Brazilian area where Chagas' disease is endemic (Dias *et al.*, 1976).

Quantitative determinations of immunoglobulin A, G and M levels were performed by single radial immunodiffusion technique (Mancini *et al.*, 1965). In our sample, these measurements were not influenced by age, sex or Chagas' disease, as revealed by multiple regression analysis (Barbosa *et al.*, 1981b).

For the study of commingling in the distribution of the immunoglobulin levels, the method of MacLean *et al.* (1976) was followed. It was assumed that the distribution of the quantitative phenotypes of a specific immunoglobulin is a commingling of either one, two or three normal distributions, whose parameters are given in Table I.

Table I - Parameters of the three different component models of the distributions.

	Component		
	1	2	3
Mean	u	$u + dt$	$u + t$
Variance	E	E	E
Proportion	$(1-q)^2 + Fq(1-q)$	$2q(1-q)(1-F)$	$q^2 + Fq(1-q)$

In the analyses, commingling is assumed to be the result of the segregation of a major gene with frequency q , the displacement t between the means of two homozygotes, the dominance effect d and the inbreeding coefficient F . A power transformation of the phenotype was used to remove skewness, so that

$$y = (t/p) [x/r + 1]^p - 1$$

where x is the immunoglobulin level, y is the transformed variable, r is a fixed scalar to insure that $(x/r)+1$ is positive ($r = 6$ was used in the present analysis) and p is the parameter of power transformation.

The sequence of the following hierarchical models was studied: the two-parameter model (u,E) was fitted and compared to the three-parameter model (u,E,p) , in order to test for skewness in the single distribution; the four-parameter model (u,E,q,t) was fitted and compared to the five parameter model (u,E,q,t,p) , in order to test for two-component commingling; the seven-parameter model (u,E,q,t,p,d,F) was fitted and compared to the five-parameter model to test for three-component commingling. When testing for one or two-component distributions, the parameter d was set to zero.

Estimation of the parameters was performed by the maximum likelihood method and hypotheses were tested using the likelihood ratio statistics. The test criterion, given by twice the difference between the log-likelihoods $(-2 \ln L + c)$ under two specific models, follows a chi-square distribution with degrees of freedom equal to the difference in the number of parameters of the two models.

RESULTS

Table II shows the descriptive statistics of the three distributions. All of them are positively skewed.

The maximum likelihood estimates of the parameters under the entertained models are given in Table III.

For IgA levels the three-parameters model fit better than the two-parameter model ($\chi^2 = 48.2$). This means that parameter p is different from zero, i.e. the

Table II - Descriptive statistics for Immunoglobulin levels in a Brazilian population, endemic for Chagas' disease.

Immunoglobulin	A	G	M
Mean (mg/100 ml)	160.03	1008.95	122.34
Standard deviation	73.99	320.13	70.80
Skewness	1.25	0.36	1.93
Kurtosis	3.18	-0.53	5.36
Sample size	302	296	292

Table III - Maximum likelihood estimates of the parameters under the three models.

Variable	Model	(-2 InL+c)	u	E	q	t	d	F	p
IgA	1 normal	426.598	-0.003	0.996	0	0	0	0	1
	1 skewed	402.520	-0.147	0.893	0	0	0	0	-0.930
	2 normal	411.947	-0.002	0.689	0.224	2.537	0	0	1
	2 skewed	402.378	-0.132	0.858	0.071	2.498	0	0	-0.724
	3 skewed	402.372	-0.128	0.820	0.068	2.700	0.215	0	-0.679
IgG	1 normal	419.504	-2x10 ⁻⁵	0.996	0	0	0	0	1
	1 skewed	414.348	-0.0867	0.991	0	0	0	0	-0.051
	2 normal	403.997	-0.0001	0.361	0.565	1.710	0	0	1
	2 skewed	403.733	-0.019	0.352	0.555	1.753	0	0	1.235
	3 skewed	403.402	-0.015	0.270	0.551	2.281	0.330	0	0.795
IgM	1 normal	413.473	-0.002	0.994	0	0	0	0	1
	1 skewed	347.217	-0.232	0.689	0	0	0	0	-2.538
	2 normal	365.226	-0.002	0.422	0.303	2.619	0	0	1
	2 skewed	344.200	-0.138	0.395	0.345	1.748	0	0	-0.998
	3 skewed	343.711	-0.159	0.244	0.288	1.445	-0.564	0	-1.338

one-component distribution is skewed. The five-parameter model fit better than the four-parameter model ($\chi^2 = 19.1$). However, there was no difference between the one-component distribution and the two-component distribution ($\chi^2 = 0.284$) and between the one-component and the three-component distributions ($\chi^2 = 0.296$). We conclude that the distribution of IgA is skewed and has only one component.

For IgG levels the three-parameter model fit better than the two-parameter model ($\chi^2 = 10.3$). No difference was found between the four and five-parameter models ($\chi^2 = 0.528$) and between the latter and the seven-parameter model ($\chi^2 = 0.662$). However, the five-parameter model fit better than the three-parameter model ($\chi^2 = 21.2$). This suggests that the distribution of IgG levels has two skewed components. As can be seen in Table III, in this case the proportion of the upper component was 0.31.

Finally, for IgM the three-parameter model fit better than the two-parameter model ($\chi^2 = 132.5$); and the five-parameter model fit better than the four-parameter model ($\chi^2 = 42.1$). There was no difference between the seven-parameter model and the

five-parameter model ($\chi^2 = 0.978$). A significant difference was found between the five-parameter model and the three-parameter model ($\chi^2 = 6.03$), which suggests that the distribution of IgM levels has two skewed components. The proportion of the upper component was 0.12.

DISCUSSION

Our analyses suggest that the distribution of IgA has one component, and those of IgG and IgM have two components. Since the effect of a major gene would lead to commingling in the distribution, our results do not support such a genetic mechanism for the regulation of IgA levels. On the other hand, for IgG and IgM the major gene hypothesis is supported.

These findings differ from those of McGue *et al.* (1989), who found significant commingling in the distribution of IgA but not in that of IgM. These authors suggested a two-component distribution for IgG and our analyses support this suggestion.

The commingling of the IgG distribution could be the result of the segregation of a major gene as well as a discrete environmental effect of a factor such as an infectious disease. A previous analysis of this sample did not reveal a significant effect of Chagas' disease on each of the immunoglobulin distributions (Barbosa *et al.*, 1981b).

The commingling in the distribution of IgM levels observed in the present study does not appear to be related to a sex-linked major gene, since we did not detect a significant effect of sex on the IgM distribution (Barbosa *et al.*, 1981b).

The differences observed in the two studies could be the result of the differences in gene frequencies and in the environmental factors between the populations.

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RESUMO

Os componentes das distribuições dos níveis séricos de IgA, IgG e IgM foram investigados em uma amostra de cerca de 300 pessoas, habitantes de uma região brasileira onde a doença de Chagas é endêmica. As distribuições de IgG e IgM parecem ser compostas de 2 componentes, ambos assimétricos; a distribuição de IgA é composta de apenas um componente. Estes resultados sugerem a ação de um gene principal ou de um evento ambiental discreto sobre as distribuições de IgG e IgM. Por sua vez, a distribuição de IgA seria determinada por componentes multifatoriais.

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