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Gm and Km Allotypes and Typhoid Fever

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Abstract

Gm and Km allotypes of immunoglobulins were determined in children with typhoid fever (Cases), in children without infectious diseases (Con-1), and in children with fever but no *Salmonella* in their blood or bone marrow (Con-2). Children were sampled from the urban population of Santiago; and they belonged to the low and low-middle socioeconomic strata. Cases had a higher frequency of [f;(–);b1,b3 or 3;(–);5,13] G1m, G2m, G3m haplotype than Con-1 and Con-2. Con-1 and Con-2 did not differ in their Gm haplotype or Km allele frequencies, but they differed in phenotype distribution. Con-1 deviated from Hardy-Weinberg equilibrium for Km due to a lack of Km 1–1 homozygotes. The relationship among these results, the ethnic origin of Chileans, and the differential susceptibility to typhoid fever are discussed.

Key Words

GM
KM
Allotypes
Typhoid fever
Case-control study

Introduction

In a previous study, we described the heterogeneous distribution of ABO, RH, MNS and sex between healthy children and children with typhoid fever [1]. This was a case-control study performed on children, aged 4–15 years, from the low-middle and low socioeconomic strata of the Northern Area of Santiago (Chile). After all laboratory determinations, serum samples from several children were still available. We typed these sera with the immunoglobulin allotype systems GM and KM and performed genetic and statistical analyses of these case-control samples.

It is important to describe some features of Chileans to envisage a possible interpretation based on ethnic admixture and differential ethnic susceptibility to disease. The present Chilean population originates from Caucasians (mostly Spaniards) and Amerindians (Mongolians), who have mixed since 1540 after being separated for more than 1,000 generations (40,000 years). Approximately 15 gen-

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erations have succeeded each other since 1540 until the present days. In the Chilean population, a sociogenetic cline was established. The highest socioeconomic strata have a small proportion of Amerindian ethnic admixture or lack it altogether, while the lowest strata show 30–50% Amerindian admixture [2]. Whereas typhoid fever had been known in Europe for more than twenty centuries ago, it only occurred after the Spanish settlement in Chile and was thus introduced from Europe [3].

Sample and Methods

The sample belongs to a case-control study on typhoid fever [1, 4]. It included three groups of children of both sexes aged 4–15 years. Controls were 80 children coming to Hospital Roberto del Río for the medical or surgical treatment of a nonfebrile process (Con-1). 'Cases' were 137 children with a febrile process coming from the outpatient clinics with typhoid fever confirmed bacteriologically by blood or bone marrow cultures of typhoid *Salmonella*. A third group was established with 378 children coming from the outpatient clinics, having a febrile process, without positive blood or bone marrow cultures of typhoid *Salmonella* (Con-2). GM allotypes were determined by the standard international hemagglutination inhibition test [5], with antisera a (1), x (2), f (3) and z (17) for G1m; n (23) for G2m; b1 (5), c3 (6), b3 (13) and g1 (21) for G3m; and 1, 3 for KM. The Gm antisera define, for G1m, the alleles [$G1m^{za}$ (17, 1)], [$G1m^{zax}$ (17, 1, 2)], [$G1m^{f}$ (3)] and [$G1m^{fa}$ (3,1)]; for G2m, the alleles [$G2m^{n+}$ (23+)] and [$G2m^{n-}$ (23-); for G3m, the alleles [$G3m^{g}$ (21)], [$G3m^{b}$ (5, 13)], [$G3m^{c3c5}$ + $G3m^{c3}$ (5, 13)] and [$G3m^{s}$ + $G3m^{st}$ (13)]. The KM antisera define two alleles [$Km^{1,2}$ + Km^{1} (1)] and [Km^{3} (3)]. Hereafter, only the numerical nomenclature will be used. The haplotypes that can be defined with these antisera are 32 (4 × 2 × 4), which define, in turn, 528 genotypes and 96 phenotypes. Maximum likelihood allele and haplotype frequencies and their standard errors, as well as deviations from Hardy-Weinberg equilibrium were obtained with the USERM1 program of the Mendel software adapted to 96 phenotypes and 528 genotypes [6]. Statistical analyis of frequencies was done by a z test of proportions, by the standard χ^2 or the likelihood ratio for the contingency tables or for the Hardy-Weinberg equilibrium.

Results

Appendix 1 presents the G1m, G2m, G3m and Km phenotypic distribution of Con-1, Con-2 and Cases among those nonempty phenotypic classes. Appendix 2 shows the distribution of nonempty extended phenotypes according to the 32 possible Gm haplotypes. A few small differences in totals were obtained because not all individuals were typed for all the systems. Table 1 shows the haplotype frequencies with their standard error for the three samples (table 1).

The frequency of [3;(–);5,13] haplotype was higher in Cases than in Con-1 ($z = 1.88$, $p < 0.03$) and Con-2 ($z = 2.43$, $p < 0.008$). There was no other significant difference in the haplotype frequencies. Con-1 and Con-2 did not differ significantly. The analysis of the phenotypic distribution among the three groups was performed by a χ^2 test for contingency of data from the Appendices. Significant differences were found between Con-1 and Con-2 for Km ($\chi^2 = 10.0$, d.f. = 2, $p < 0.007$) due to a higher frequency of Km 1–3 in Con-1; and between Con-1 and Con-2 for Gm ($\chi^2 = 34.1$, d.f. = 22, $p < 0.049$) due to a higher frequency of phenotypes (17,1,3,23,5,13), (17,1,2,21,13), and (17,1,23,21,5,13) in Con-1. However, these last two phenotypes were found in 2 and 1 individuals, respectively, in Con-1, whereas they were detected in none of the individuals in Con-2.

Only Con-1 showed a significant deviation from Hardy-Weinberg equilibrium in Km due to a lack of homozygotes 1–1 ($p = 0.002$). Con-2 and Cases did not differ from the panmictic equilibrium in Km. G1m and G2m did not deviate from Hardy-Weinberg equilibrium in any sample.

Table 1. Haplotype and allele frequencies in Con-1, Con-2 and cases

Haplotype			Con-1 (n = 80) frequency	SE	Con-2 (n = 378) frequency	SE	Cases (n = 137) frequency	SE
G1	G2	G3						
Gm								
17,1	23	21	0.02752	0.01872	0.01722	0.00690	0.00806	0.07984
17,1	23	5,13	0.01006	0.01376	0.00000	0.00000	0.01159	0.01335
17,1,2	23	21	0.01103	0.01383	0.00490	0.00406	0.01219	0.00855
3	23	5,13	0.23680	0.03613	0.25510	0.01681	0.21190	0.02754
17,1	(–)	21	0.41710	0.04170	0.45830	0.01891	0.44150	0.03101
17,1	(–)	5,13	0.07119	0.02324	0.03766	0.00699	0.02876	0.01724
17,1	(–)	5,6	0.00000	0.00000	0.00396	0.00229	0.00000	0.00000
17,1,2	(–)	21	0.06931	0.02385	0.06784	0.00984	0.05965	0.01627
17,1,2	(–)	13	0.01875	0.01073	0.00000	0.00000	0.00024	0.00438
3	(–)	21	0.00000	0.00000	0.01541	0.00501	0.01029	0.00716
3	(–)	5,13	0.13810	0.03033	0.13950	0.01402	0.21570	0.02800
Km	Allele		Con-1 (n = 82)	SE	Con-2 (n = 378)	SE	Cases (n = 138)	SE
	1		0.28660	0.03531	0.22880	0.01528	0.27170	0.02678
	3		0.71340	0.03531	0.77120	0.01528	0.72830	0.02678

Discussion

In this study we are sure that Cases are susceptible to typhoid fever. Con-1 individuals had no episode of infection near the moment of ascertainment. Con-2 had febrile episodes, but no *Salmonella* in their blood or bone marrow. Individuals carrying the 3;(–);5,13 haplotype were found to be more susceptible to typhoid fever (Cases). The frequency of this haplotype in Cases was higher than in Con-1 ($p < 0.03$) and Con-2 ($p < 0.008$). This result can hardly be attributed to chance alone. Since there were 11 possible comparisons per sample, the true significance could be less than that found. However, the corrected maximal probability should be $p = 11 \times 0.03 \times 11 \times 0.008 = 0.02904$, which is still significant. This haplotype is more frequent in Caucasians (0.249) than in Amerindians (0.0), the founder groups of Chileans [unpubl. data and 7]. Since thyphoid *Salmonella* was introduced in Chile by Spaniards that might have been adapted to them, it is difficult to explain the higher susceptibility to typhoid fever of children with this haplotype. Perhaps, it is a fortuitous association or Spaniards who came to Chile really had susceptibility associated to this haplotype. This association might also have arisen in the 15 generations after the original mixture.

Con-1 and Con-2 did not differ in allele and haplotype frequencies, but they differed in phenotype distribution. This could be due to a different gene interaction between alleles in febrile and nonfebrile individuals. An allele or haplotype could induce a different susceptibility to febrile processes depending on the different complete genotypes. The number of individuals with phenotypes 17,1,3,23,5,13 and Km 3–3 was sufficient for a valid comparison. Thus, the difference between the two

samples is probably due to a true biotic condition.

Only Km, in Con-1, showed a deviation from Hardy-Weinberg equilibrium due to a lack of 1–1 and an excess of 1–3 individuals. The deviation remains significant considering the 9 independent tests for Hardy-Weinberg equilibrium ($0.002 \times 9 = 0.018$). Since Con-1 are nonfebrile controls, it is difficult to explain such a deviation. Deviations from Hardy-Weinberg equilibrium have been found in other healthy control samples [1, 8].

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Appendix 1

G1m, G2m, G3m and Km phenotype distribution in Con-1, Con-2 and cases

Phenotype		Con-1	Con-2	Cases
G1m	17,1	20	93	30
	17,1,2	9	31	11
	17,1,3	36	176	66
	17,1,2,3	6	22	8
	3	9	56	23
	3,1	0	0	0
	Total	80	378	138
G2m	23	39	184	60
	(–)	43	194	78
	Total	82	378	138
G3m	21	21	115	39
	21,5,13	39	194	67
	21,13	2	0	0
	5,13	17	66	31
	5,6,13	2	3	0
	13	1	0	0
	Total	82	378	137
Km	1–1	1	17	6
	1–3	45	139	63
	3–3	36	222	69
	Total	82	378	138

Appendix 2

Distribution of phenotypes from extended Gm haplotypes in Con-1, Con-2 and cases

Phenotype	Con-1	Con-2	Cases
17,1,23,21	2	6	1
17,1,23,21,5,13	1	0	0
17,1,2,23,21	1	3	2
17,1,3,23,21,5,13	18	108	29
17,1,3,23,5,13	5	6	7
17,1,3,23,5,6,13	0	1	0
17,1,2,3,23,21,5,13	5	16	5
17,1,2,3,23,5,13	1	0	0
3,23,21,5,13	0	1	0
3,23,5,13	7	43	15
17,1,21	12	73	26
17,1,21,5,13	4	11	2
17,1,2,21	6	25	8
17,1,2,21,5,13	0	3	1
17,1,2,21,13	2	0	0
17,1,3,21	0	4	1
17,1,3,21,5,13	12	53	28
17,1,5,13	1	1	0
17,1,5,6,13	0	2	0
17,1,3,5,13	1	4	1
17,1,2,3,21	0	4	1
17,1,2,3,21,5,13	0	2	2
3,5,13	2	12	8
Total	80	378	137

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