

# Clustering of malformations in the families of South American oral cleft neonates

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## Abstract

The relatives of 741 newborn children with non-syndromic cleft lip with or without cleft palate (CL±P), of 115 with isolated cleft palate (CP), and of equal numbers of appropriate controls were screened for the presence of the same or different malformations. The main findings were as follows. (1) The frequency of familial cases of CL±P (17.3%) was much higher than the prevalence of this malformation among the relatives of controls (0.5%). (2) The sibs of CL±P subjects showed a higher prevalence of this condition than their parents (2.9% *v* 1.6%). (3) The degree of genetic determination of this condition should be high (70 to 74%), and the data in general favour a multifactorial model of inheritance, with different thresholds between sexes. However, the action of dominant genes cannot be excluded since selection or dominant genes or both could be postulated to explain the parent/sib difference. (4) The frequency of other malformations was also significantly raised in the families of CL±P probands, as compared to controls (12.1% *v* 6.2%). (5) The prevalence of these other malformations was higher among sibs (1.6%) than parents (0.7%) of CL±P babies. (6) A general susceptibility to malformations and different exposure to selective agents may explain these latter findings. (7) None of the comparisons involving CP children yielded significant results.

The fact that cleft lip with or without cleft palate (CL±P) and isolated cleft palate (CP) occur more frequently in the families of subjects with these malformations than in the general population is well known, the independence between these entities having been established in the 1940s.<sup>1</sup> Recent reviews

on the genetics of these conditions can be found in Bear<sup>2</sup> and Fraser.<sup>3</sup> However, the causes of liability to these conditions are far from clear, especially the variation in risk according to the type of proband considered (whether they are males or females, and have a more or less severe malformation), a question that is vital for the establishment of the appropriate model of disease liability.<sup>4-7</sup> Estimates of the degree of genetic determination of these entities have also been quite variable.<sup>5-12</sup> Another problem is the relationship between the susceptibility to these conditions and to other malformations, excluding the syndromes of which CL±P and CP are a part. Here again, there are conflicting results and interpretations.<sup>12-14</sup> Therefore, we decided to investigate certain aspects of these questions as part of a larger study, which included an epidemiological search and the analysis of the association between these anomalies and fetal death.<sup>15 16</sup>

## Subjects and methods

The data were obtained through the Latin American Study of Congenital Malformations (ECLAMC, Estudio Colaborativo Latinoamericano de Malformaciones Congénitas). This is a clinical-epidemiological programme of the case control type, based in hospitals, the aim of which is the investigation of congenital defects detected in the neonatal period. This study has been operating in several South American countries since 1967. All babies born in the hospitals which participate in the programme (at present there are 82 hospitals, located in 10 countries) are examined at birth by a trained paediatrician, and minor as well as severe malformations are described in detail. Information about risk factors and family histories are obtained by this physician directly from the mother in the postpartum period. Each malformed child is allocated a control, which is the first normal baby of the same sex born in that hospital after the malformed one.

All cases of CL±P and of CP ascertained during the period 1967 to 1981 who had no associated anomalies were selected for the study, and the data were transferred to the files of a PC-XT microcomputer, using the dbase III plus program. Subsequently, they were subjected to standard statistical procedures.

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Table 1 Prevalence of the same or different types of malformation among the relatives of probands and controls.

Degree of relationship	Relatives with CL±P*		Relatives with CP*		Relatives with other malformations*			
	CL±P	Control	CP	Control	CL±P	Control	CP	Control
1/2 (parents)	24	0	0	0	10	3	1	0
(sibs)	37†	0	1	0	20	9	3	1
1/4	34	1	1	0	28	21	2	2
1/8	35	4	1	0	21	14	3	2
1/16	19	2	0	0	10	2	1	1
1/32	4	0	0	0	1	1	0	0
<1/32	3	0	0	0	1	0	0	0
Unspecified	3	0	0	0	24	9	4	2
Total	159	7	3	0	115	59	14	8
No of families	741	741	115	115	741	741	115	115
No of families with affected relatives	127‡	4	3	0	90	46	12	8
% of families with affected relatives	17.3	0.5	2.6	0.0	12.1	6.2	10.4	7.0

\*Information about total number of relatives was available for parents and sibs only. They are as follows. (1) Parents: CL±P cases and controls, 1480; CP cases and controls, 230. (2) Sibs: CL±P cases, 1270; controls, 1003; CP cases 170, controls, 138.

†32 sibships with one affected, one with two affected, and one with three affected, besides the proband.

‡One family with two probands (twins).

Type and degree of genetic determination were estimated according to Czeizel and Tusnady<sup>12</sup> and Emery.<sup>17</sup> Further details about this sample have been presented elsewhere.<sup>15 16</sup>

## Results

Table 1 shows the prevalences of the same type or of different types of malformation among relatives of the probands and those of appropriate controls. The following points should be stressed. (1) The frequency of familial cases of CL±P was 17.3%, compared to 0.5% of subjects with this malformation among the families of controls. The corresponding figures for CP show a less marked difference (2.6% *v* 0.0%). (2) If only subjects who share 50% of their genes with the probands are included, it will be seen that the frequency of CL±P among the sibs of newborn babies with this malformation ( $37 \times 100 / 1270 = 2.9\%$ ) is significantly higher than that present among their parents ( $24 \times 100 / 1480 = 1.6\%$ ;  $p < 0.02$ ). (3) The frequency of other malformations is also significantly raised in the families of CL±P probands, as compared to those of controls (12.1% *v* 6.2%;  $p < 0.02$ ), but not among the families of CP probands (10.4 *v* 7.0%). (4) The prevalence of other malformations is higher ( $p < 0.05$ ) among the sibs ( $20 \times 100 / 1270 = 1.6\%$ ) than the parents ( $10 \times 100 / 1480 = 0.7\%$ ) of CL±P probands.

Additional analyses can be made in the larger CL±P series, but even there sample sizes prevent separate analyses of parents and sibs. Therefore, they are grouped together as first degree relatives, as other authors have done; the results are given in table 2. The difference in frequency between the prevalence of this malformation among first degree relatives of CL±P male (1.8%) and female (2.8%) probands is as expected for a model of multifactorial inheritance with different thresholds for the manifestation of the

Table 2 Prevalence of the same malformation among first degree relatives of different types of CL±P probands.

Types of CL±P probands	First degree relatives			
	No	Affected	Total	Percentage
Males	447	29	1591	1.8
Females	294	32	1159	2.8
Total	741	61	2750	2.2
CL only	236	17	841	2.0
CL+P	505	44	1909	2.3
Unilateral CL	201	15	723	2.1
Bilateral CL	17	4	53	7.5
Unilateral CL+P	332	26	1305	2.0
Bilateral CL+P	132	12	308	3.9

For 59 probands there was no information on whether the malformation was uni- or bilateral.

malformation in the two sexes, but the difference is not significant. The comparison of prevalence between the relatives of probands with CL only and CL+P also yielded non-significant results, but the relatives of unilateral CL had a lower prevalence of the malformation (2.1%) than those of bilateral CL cases (7.5%;  $p < 0.01$ ), the same being true when unilateral and bilateral CL+P probands are considered (2.0% *v* 3.9%;  $p < 0.05$ ).

A search was conducted to verify if, among the other malformations present in the relatives of CL±P or CP babies, the head region was more often affected than other parts of the body. However, this was not found to be the case. Neural tube or other fusion defects were not particularly frequent either.

The degree of genetic determination of CL±P can be estimated by comparing the frequency of the malformation among first degree relatives of the probands (2.2%) with that obtained in the total sample (0.87).<sup>15</sup> Using a graph constructed by C A B Smith and reproduced in Emery,<sup>17</sup> which considers the relationships between these frequencies and the

parameter considered, we arrived at an estimate of 70%. It is also possible to calculate the degree of genetic determination using another method,<sup>12</sup> but the value obtained (74%) is similar to that reached by the first method. By dividing the prevalence among first degree relatives by that found in the general population we obtain a value (named K by Penrose in 1953<sup>17</sup>) which can serve as a rough indicator of the mode of inheritance of a given characteristic. For CL±P the number obtained was 33·3, almost identical to that expected under a model of multifactorial inheritance (33·9; the number expected for an autosomal dominant gene would be much higher: 574·7).

For cleft palate alone the numbers do not justify the performance of such calculations. Taken at face value, however, the presence of one affected by the same malformation among the first degree relatives of the CP probands yields a frequency of 1×1000/230 parents+170 sibs=2·5‰, 19·2 times higher than the population prevalence of 0·13‰.<sup>15</sup>

### Discussion

The estimates of the degree of genetic determination obtained here for CL±P (70 to 74%) are very close to those found in two European countries, in the United States, and in China (74 to 79%; comparison restricted to samples in which more than one category of relatives was compared with the general population).<sup>5 6 8 9 11</sup> This similarity, observed in such variable environments, suggests that the real figure should be within the interval provided by these studies.

As for the differences found in the prevalences of CL±P in relatives of different CL±P probands, table 3 compares the results reported here with those obtained previously in different studies. In all cases the present figures are within the range of those encountered in earlier surveys, although it should be mentioned that sometimes this range is quite wide.

Table 3 Prevalence of CL±P among first degree relatives of different types of CL±P probands: comparison among series.

Types of CL±P probands	Frequency in first degree relatives (%)			
	Present study	Previous studies <sup>4-7</sup>		
		No of series	Average	Range
Males	1·8	5	2·8	1·2-4·0
Females	2·8	5	3·1	1·9-3·8
CL only	2·0	7	2·6	2·0-4·3
CL+P	2·3	7	2·9	1·3-3·9
Unilateral CL	2·1	5	2·2	1·0-2·7
Bilateral CL	7·5	3	4·0	0·0-8·3
Unilateral CL+P	2·0	5	3·4	1·3-4·2
Bilateral CL+P	3·9	5	4·7	1·4-6·6

The lack of difference in the number of affected relatives of CL as compared to CL+P subjects has also been found elsewhere, the averages of these previous investigations (2·6 v 2·9) being similar to those reported here (2·0 v 2·3). This is evidence against the multifactorial model, but all the other comparisons favour it, as well as the different thresholds of males and females. The recently reported association between two restriction fragment length polymorphisms at the transforming growth factor alpha locus and the occurrence of CL±P<sup>18</sup> is interesting in this regard. Confirmation or refutation of this finding may be decisive in the search for the elusive major gene that could influence the liability to this complex malformation. Other recent analyses<sup>19 20</sup> suggest that the effects of a major gene should not be ignored.

We have found that the frequency of CL±P is higher among sibs (2·9%) as compared to parents (1·6%) of CL±P probands. Similar results were observed in the majority of previous studies,<sup>5-7 12 21 22</sup> but there are two conflicting reports.<sup>23 24</sup> Two explanations can be advanced for this difference: (1) reduced viability of CL±P subjects, or (2) action of dominant genes in the background of such subjects.<sup>25</sup>

The prevalence of other malformations was two times higher in the families of CL±P probands than in those of the controls. This was not observed by Czeizel and Tusnady,<sup>12</sup> who found similar frequencies in these two groups. On the other hand, Fraser *et al*<sup>14</sup> encountered a high frequency of neural tube defects among the sibs of CL±P subjects, and Khoury *et al*<sup>13</sup> observed a high incidence of CL±P in the sibships of probands with neural tube defects without other associated malformations. Both anomalies are more commonly found in spontaneous abortions than in livebirths. Therefore, the association could result from a diminished capacity of certain mothers to reject determined types of malformed embryos, or, alternatively, from a uterine, familial, environmental, or genetic embryonic factor that would increase the probability of various fusion defects.<sup>14</sup> It should be noted, however, that in our series neural tube or other fusion defects were not overrepresented. We are left, therefore, with two other explanations for the findings reported here: (1) a general susceptibility to malformations in the families of CL±P subjects; or (2) differing recall among the mothers of affected babies compared to those of normal babies. Against the second alternative is the fact that no significant differences regarding these frequencies were found in the families of CP and normal newborn children.

Finally, the two times higher prevalence of other malformations among sibs as compared to parents of CL±P children could be explained by the fact that the former had not yet been completely subjected to the action of natural selection, as had the subjects of the earlier generation.

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