CLINICAL STUDY

Coxsackie viral infection and orofacial cleft

Molnarova A, Petrovicova A, Fedeles J, Bopegamage S, Horakova E

Plastic Surgery Department, Faculty of Medicine Comenius University, University Hospital Ruzinov, Bratislava, Slovakia. **petrovic@upkm.sk**

Abstract

The incidence of orofacial cleft (OC) in newborns was compared with the occurrence of virus-neutralizing antibodies to coxsackie viruses in the serum of newborns and their mothers. No significant difference was found when comparing the seropositivity rates between the group of patients and the control group of healthy newborns. If the patients were divided according to the place of residence however, marked differences occurred between the regions. The lowest incidence of both – coxsackie infection and OC was determined in the region of Bratislava and the highest in the region of Žilina. The explanation of these findings recquires a more detailed analysis of genetic background, social and hygienic status, style of life and other factors, known to influence the development of OC as multi-etiological developmental disorder. ***(Tab. 2, Fig. 4, Ref. 12.)***

Key words: orofacial cleft, coxsackie-virus infection, regional distribution of malformations.

Orofacial cleft (OC) is a very important and one of the most often found congenital malformations. It develops during 5th–12th weeks of pregnancy. The aetiology of OC is complex and heterogeneous both for isolated and associated defects. Causes linked to environment, genetics and gene-environment interaction are known, although a lot work is required to make the events clear (1). The genetic basis is connected with variation in multiple genes (2). In the pathogenesis interaction of the above-mentioned genetic and so-called environmental factors, as stress (3), alcohol consumption (4, 5), mother's nutritional status (6) and viral infections (7) is supposed.

Many viruses can infect the foetus through transplacental or (less frequently) ascendent routes. Connection between coxsackie viral infection and congenital malformations is expected, but the exact evidence is still missing. The ability of coxsackie viruses to infect placenta has been proved in different studies (8, 9). Foetal infection with coxsackie-viruses B2 and B6 was demonstrated in spontaneous abortions (8). Anticoxsackie viral antibodies were detected in the ventricular fluid of newborns with CNS malformations (9). Experimental studies showed, that infection of pregnant mice with coxsackie-viruses causes defects in the foetoprotein production with subsequent retardation and disorders of the foetus growth (10). Detection of coxsackie viruses using PCR in the placental tissue of newborns with neurodevelopmental delays was positive in 85 % of cases with an absence of positivity in control healthy group (11).

Material nad methods

The markers of outdated coxsackie-viral infection (presence of antibodies against 8 types of coxsackie B viruses) were followed in serum samples from 189 newborns with OC and their mothers (Group 1) and compared with those markers in control group of 62 healthy newborns and their mothers (Group 2). The samples were collected in the Plastic Surgery Department, Medical School UC, Bratislava during 1991–2001. The antibody titres were determined in micromodification of virus-neutralisation test in cell cultures (green monkey kidney cells – GMK). In this study the anticoxsackie-viral antibodies of IgM class were not assayed, because the OC (according to the literature data) develops during 5th–12th weeks of pregnancy, therefore at the time of birth in the mother's sera are IgM antibodies connected with acute coxsackie viral infection in over-mentioned interval of pregnancy not detectable (outlast 8 weeks in detectable levels). In addition to it, nowadays no commercial diagnostic set

Plastic Surgery Department, Faculty of Medicine, University Hospital Ruzinov, Bratislava, Institute of Preventive and Clinical Medicine, Bratislava, and 2nd Gynaecological Department, Faculty of Medicine, University Hospital Ruzinov, Bratislava

Address for correspondence: A. Petrovicova, MD, PhD, UPKM, Limbova 14, SK-833 01 Bratislava, Slovakia.
Phone: +421.2.59369555, Fax: +421.2.54773906

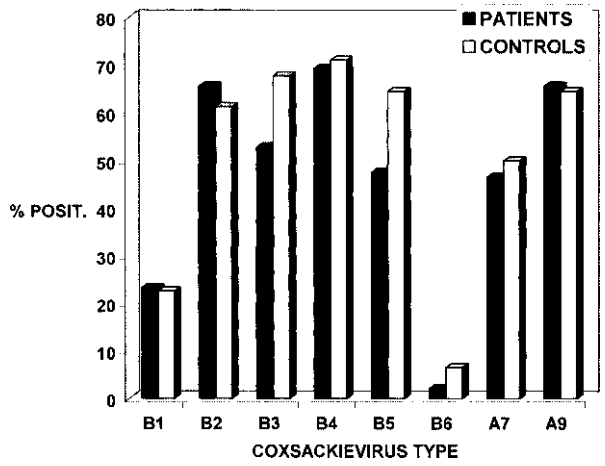

Fig. 1 a. Presence of antibodies against 8 coxsackie-virus serotypes in sera of newborns with OC (patients) and healthy newborns (controls).

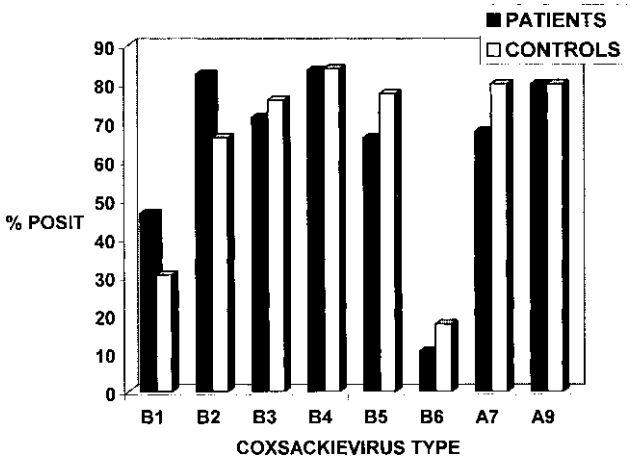

Fig. 1 b. Presence of antibodies against 8 coxsackie-virus serotypes in sera of newborns with OC (patients) and healthy newborns (controls).

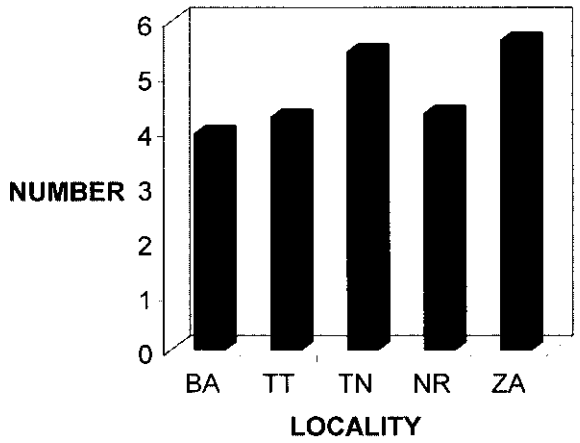

Fig. 2 a. Mean number of coxsackie-virus serotypes against which antibodies were present in sera of mothers of newborns with OC according to the regions of place of residence. BA — Bratislava, TT — Trnava, TN — Trenčín, NR - Nitra, ZA — Žilina.

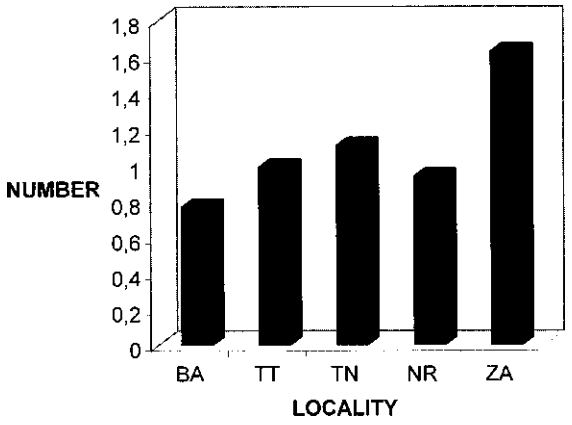

Fig. 2 b. Annual incidence of OC in newborns in different regions of Slovakia. BA — Bratislava, TT — Trnava, TN — Trenčín, NR — Nitra, ZA — Žilina.

for anticoxsackie viral IgM antibodies detection is available and the "in-house" method used in our laboratory for diagnostic purposes is too laborious and costly and requires large amounts of serum. Therefore for this preliminary study we choose to assay only whole anticoxsackie viral antibodies. Statistical significance was calculated using chi^2 test.

Results and discussion

The presence of antibodies against different coxsackie viruses in mother's sera from Group 1 and Group 2 show no significant differences (Figs 1a and 1b). The role of repeated coxsackie viral infection on the manifestation of genetical disposition to the development of OC was tested via comparison of the number of coxsackie viral types against which specific antibodies were detected in mothers' sera (Tab. 1). But no statistically significant differences between Group 1 and Group 2 were found.

Then the patients were divided according to the place of residence into regions Bratislava (BA), Trnava (TT), Trenčín (TN), Nitra (NR), Žilina (ZA) and common region Banská Bystrica, Prešov and Košice (BPK). Because of the low number of cases in the region BPK in our study group (due the the fact, that patients from this region belong to the hinterland of the Plastic Surgery Department in Košice and only occasionally are hospitalised in Bratislava), further comparison was done only in patients living in 5 regions (BA, TT, TN, NR, ZA). The same mark-

Tab. 1. The seropositivity against coxsackie virus serotypes detected in the sera of mothers – the mean number of serotypes (M).

Group	M
Patients	5.08
Controls	5.02

M — mean number of coxsackie virus serotypes against which antibodies were detected in the serum samples

Tab. 2. The seropositivity against coxsackie virus serotypes detected in the sera of mothers and the annual incidence of OC according the place of residence.

Locality	M	Annual incidence of OC
BA	3.95	0.77
TT	4.25	0.99
TN	5.44	1.11
NR	4.32	0.94
ZA	5.65	1.63

Locality — region of the place of residence, BA — Bratislava, TT — Trnava, TN — Trenčín, NR — Nitra, ZA — Žilina
M — mean number of coxsackie-virus serotypes against which antibodies were detected in the serum samples

ers like in the Table 1 were compared with the annual incidence of OC in the relevant region. The results presented in Table 2 and Figs 2a and 2b show correlation between the annual incidence of OC (per 100,000 inhabitants in given county) and number of coxsackie virus serotypes, against which antibodies were detected. In the BA region was the lowest incidence of OC and the lowest mean number of serotypes against which antibodies were detected, while in ZA region both values were the highest. Detailed explanation of the finding at the time is not within reach, but differences between the social and hygienic levels, nutrition and style of life between these regions are known. All of them can influence the frequency of enteroviral infections and incidence of OC in the given population.

These data as the results of a preliminary study support the hypothesis, that repeated viral infection can act as trigger of genetic predisposition, but further more detailed studies are required to confirm the presumption.

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