Perinatal cytomegalovirus infection in man

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SUMMARY In a prospective study of 148 children from urbanized southern Finland 3 were found to be congenitally and 48 perinatally infected with cytomegalovirus (CMV), while 6 developed 'late' infection during the first year of life. During pregnancy and the first year after delivery 23 of the mothers had no CMV antibodies; none of the children of these seronegative mothers developed any type of CMV infection. Fresh blood exchange transfusions did not increase the risk of CMV infection. The data support the hypothesis that the mother is the source of perinatal CMV infection. Children with a low birthweight not due to prematurity, and first children seem to run a greater risk of acquiring perinatal CMV infection. If the child is breast fed up to the age of 2 months the risk seems to be increased. Perinatal CMV infection gave rise to no symptoms or signs and had no effect on growth or on motor and psychosocial development during the first year of life.

Cytomegalovirus causes intrauterine infection in 0.7 to 1.8% of all children and several follow-up studies have shown a high frequency of at least mild developmental disorders of the central nervous system among congenitally infected infants (Starr *et al.*, 1970; Melish and Hanshaw, 1973; Stern and Tucker, 1973; Reynolds *et al.*, 1974). Some congenital infections are symptomless, and at birth only a few congenitally infected babies show typical severe cytomegalic inclusion body disease (CID) with hepatosplenomegaly, thrombocytopenia, seizures, chorioretinitis, etc. (Starr *et al.*, 1970; Hanshaw, 1971). Previous investigations also indicate that CMV infection is very common in the first months of life. This type of infection has been called perinatal (Levinsohn *et al.*, 1969; Numazaki *et al.*, 1970; Reynolds *et al.*, 1973).

CMV has been isolated from the uterine cervix most frequently towards the end of pregnancy, and many authors have suggested that cervical virus may be responsible for perinatal infection (Numazaki *et al.*, 1970; Montgomery *et al.*, 1972; Reynolds *et al.*, 1973). It has been shown that perinatal CMV infection is asymptomatic and, like congenital infection, leads to virus excretion persisting for several months or even years in spite of appreciable levels of circulating antibody (Numazaki *et al.*, 1970; Reynolds *et al.*, 1973). The question has been raised as to whether perinatal infection may lead to later sequelae involving the central nervous system as does congenital infection (Reynolds *et al.*, 1973; Kibrick and Loria, 1974).

In order to find out whether perinatal infection as such may cause clinical signs or symptoms which might be recorded during the first year of life as clinical acute infection or developmental disorder, a careful follow-up comparison was made between 48 perinatally infected and 91 noninfected infants. These belonged to the material of a prospective follow-up study originally consisting of 200 mothers and babies. In addition, 3 congenitally infected infants and 6 who had acquired CMV infection after 6 months of age were included in this study. This series also offered a good opportunity for evaluating the predisposing factors which increase the risk of CMV infection during the first few months of life.

Material and methods

Collection of series. Women attending the Obstetric Department of the University Central Hospital, Helsinki, from 6 September to 31 October 1972, for delivery were asked to participate in this study. Only women who had been living in or close to Helsinki during their pregnancy were asked, since in this area a serum sample had been collected from all mothers during their early pregnancy in connection with another study (Lapinleimu *et al.*, 1974).

200 mother-child pairs were thus chosen to participate. The hospital records were used for a retrospective follow-up of pregnancy and delivery. The socioeconomic backgrounds were registered by means of a questionnaire. Of the original 200 infants, 148 could be followed throughout the first

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year of life. Virus isolation and antibody assay methods have been described in detail elsewhere (Leinikki, 1972; Leinikki *et al.*, 1972).

Clinical signs. The infants had a thorough clinical examination at the ages of 2, 4, 6, 8, and 12 months. Virological samples were collected at the same time. A physical examination was carried out; height, weight, and head circumference were recorded; and the developmental state was assessed. Further tests were carried out if signs of illness were noted. The mothers were asked at each visit about the occurrence of any signs of illness. A nurse was available daily for consultation and advice if required. 2 infants died during the first days of life, 1 of a severe heart malformation and the other of a septic illness. 3 severely handicapped infants were also lost from the study. None of these 5 was congenitally infected with CMV, and none of the 3 handicapped infants had had a perinatal infection.

Results

Epidemiology. During the first year of life 39% of all infants started to excrete CMV in their urine. Virus excretion at various ages is shown in Table 1.

Table 1 *Number of infants (total=148) who had started excreting virus by age groups*

Age (m)	No.	%
0	3	2
2	24	16
4	48	32
6	51	36
8	55	37
12	57	39

3 congenitally infected infants excreted virus during the first days of life. At the age of 4 months 32% were excretors. Infants who began to excrete virus at the age of 2–5 months were considered to be perinatally infected. A few were apparently infected later, virus excretion beginning after the age of 6 months ('late' infection) (Table 2).

CMV antibody assays performed during pregnancy, at delivery, and later proved that 23 mothers were seronegative (lacking CMV antibodies). Their

children showed neither congenital, perinatal, nor 'late' CMV-infection. CMV antibody assays also showed increased titres at the same age as the virus excretion began. The antibody levels of the congenitally infected infants were stable during the first year of life, and the noninfected infants slowly lost their maternal antibodies during the first 6 months.

Factors related to pregnancy, birth, and physical and psychosocial development could only be compared in the perinatally infected and non-infected children because the other groups were too small.

Comparison of CMV infected and noninfected children with regard to factors related to pregnancy and birth. No significant differences were observed in the mean relative weight of the placenta, mean age of the mothers, mean birthweight and length, or mean head circumference at birth (Table 3). The duration of pregnancy was 38–41 weeks in 87% of cases. The distribution of unusual gestational ages, relative weights of the placenta, and ages of the mothers, as well as the frequency of deliveries by caesarean section showed no significant differences (Table 4). Though not statistically significant, low birthweight and perinatal CMV infection seemed to go in parallel, especially among girls (Fig. 1). 7 out of 10 girls with a birthweight under 3000 g were perinatally infected.

The one-minute Apgar scores of 146 infants ranged from 6 to 10 (Fig. 2). The score dropped after one minute in only one case. The only difference between the perinatally infected and noninfected infants occurred at Apgar scores of 8 and 9. A score of 9 was more common among the noninfected than among the perinatally infected group but not significantly so.

Because of threatening asphyxia or malpresentation 8 babies were delivered by caesarean section. 3 of these developed perinatal infection in spite of intact fetal membranes. Another developed 'late' infection. 3 of those 4 infants who did not acquire CMV infection had ruptured membranes at section. The time of rupture varied from 2 to 24 hours before delivery.

Table 2 *Distribution of infants by type of infection*

Type of CMV infection	No.	%	Boys/girls
Congenital	3	2.0	0/3
Perinatal	48	32.5	23/25
'Late'	6	4.1	3/3
Not infected	91 (23)*	61.4 (15.3)*	51/40 (17/6)*
Total	148	100.0	77/71

*Children of CMV seronegative mothers.

Table 3 Mean values for birthweight, height, and head circumference, the relative weights of the placenta, and the mean age of the mothers in various infection groups

	All	Infection group		
		Perinatal	Congenital	Noninfected
Birthweight (g)				
Boys	3583	3699		3516
Girls	3580	3461	3570	3557
Birth length (cm)				
Boys	50.7	51.2		50.5
Girls	50.2	49.9	50.3	50.2
Head circumference (cm)				
Boys	35.3	35.6		35.1
Girls	35.0	34.7	35.5	35.2
Relative weight of placenta (%)				
Boys	18	17		18
Girls	19	18	18	19
Age of mothers				
Boys	26.4	25.7		26.7
Girls	26.8	25.4	22.3	27.4

Table 4 Distribution of unusual gestational ages and relative weights of the placenta, deliveries by caesarean section, and occurrence of old and young mothers in various infection group

	Infection groups				All	
	Congenital n=3	Perinatal n=48	Late n=6	Noninfected n=91	n=148	%
Gestational age						
>41 w		3	1	6	10	6.8
<38 w		3		6	9	6.1
Relative weight of the placenta						
>21%	1	5		9	15	10.1
<15%		4		8	12	8.1
Age of mother						
<20 yr	1	2	1	3	7	4.7
>35 yr		1	1	7	9	6.1
Delivery by caesarean section		3	1	4	8	5.4

Fig. 1 Numbers of perinatally infected and noninfected infants in various birthweight groups.

Fig. 2 Distribution of Apgar scores among noninfected and perinatally infected infants.

58% of the infants were born as the first child, 30% as the second, and 10% as the third. First-born children were relatively more common in the perinatally infected than in the noninfected group. Comparing only those infants who were exposed

to CMV infection from their (seropositive) mothers, the difference was more apparent (Fig. 3).

Fourteen infants (9%) were hospitalized during the first week of life, 7 for hyperbilirubinaemia. 6 of the latter received exchange transfusions of fresh blood. 2 of these infants were perinatally infected and began to excrete CMV in their urine at the age of 2 months. Perinatal infection was also manifested at the age of 2 months in a girl with spherocytic anaemia who received both fresh and restored blood. One of the infants treated by transfusion of fresh blood was born to a seronegative mother and remained free from CMV infection. 4 cases were hospitalized for suspected infection, 1 of whom had begun to excrete CMV at the age of 2 months.

Fig. 3 Birth order of the children of seropositive mothers (only perinatally infected and noninfected infants included).

Follow-up during the first year of life. Physical development was similar in all groups. The increase in weight, height, and head circumference followed Finnish standards closely (Bäckström and Kantero, 1971; Kantero and Tiisala, 1971). Analysis of psychomotor development showed no abnormalities. At the last examination 75 infants could walk and 100 could speak at least one word. Because the ages varied from 11 to 13 months at this examination, only those infants who were 12 months $\pm$ 1 week old on these occasions were included in a more detailed analysis. 31 out of 44 (71%) of the noninfected infants and 18 out of 26 (69%) of the perinatally infected infants could speak, and 23 (52%) of the former and 15 (57.6%) of the latter could walk. 2 of the 3 congenitally infected children attended this examination. Both of them could walk and speak at the age of 12 months.

Since breast milk is one potential source of CMV transmission from mother to child, the duration of breast feeding was registered in all cases. In 72 it

was discontinued before the age of 2 months, and only 18 infants were breast fed for more than 4 months. At the age of 2 months the perinatally infected infants were significantly more often breast fed than noninfected infants ($P < 0.01$, critical ratio) (Fig. 4) (33 = 69% of the perinatally infected, and 37 = 41% of noninfected infants, respectively). However, by the age of 4 months this difference had almost disappeared. A few infants only were not breast fed and therefore a comparison of breast-fed and nonbreast-fed infants could not be made.

Fig. 4 Number of breast-fed infants of various ages. Noninfected and perinatally infected included.

Respiratory infection was the most frequent clinical illness after the neonatal period, and then gastroenteritis, rashes, and urinary infection. For the 15 cases hospitalized the outcome was favourable in all and both the severity and distribution of infection were similar in the perinatally infected and noninfected groups. Every infant on average had three clinically manifested infectious episodes during the first year of life. Illnesses were rare during the first 2 to 3 months, though these infants were born in the autumn. No signs or symptoms could be related to the perinatal infection. The only exceptions were 3 perinatally infected girls, who developed urinary tract infection at 4 to 5 months of age.

Discussion

Perinatal CMV infection is very common. In the present study 32% of all children and 38% of the children of seropositive mothers started to excrete virus before the age of 4 months. A few acquired CMV infection after this age. The risk of acquiring a CMV infection is evidently greatest during the

first months of life, and this agrees with earlier reports (Levinsohn *et al.*, 1969; Numazaki *et al.*, 1970; Leinikki *et al.*, 1972; Stoll, 1974). The pattern of perinatal CMV infection seems to be uniform with regard to epidemiology and probably with regard to mode of infection. This type of CMV infection, moreover, seems to be the usual epidemiological route of perpetuation of the virus.

The importance of the mother as the source of infection is obvious, since none of the children of seronegative mothers developed CMV infection during the first year of life. All these 23 infants had a good opportunity to become infected during the examinations, being in close contact with other children in the examination and waiting rooms. The mechanism of transmission of the virus from mother to child probably implies the activation of a latent CMV infection in the mother during pregnancy. The transmission of CMV from mother to child may take place before birth, in the birth canal during delivery or after birth, since CMV is very often found in the uterine cervix (Numazaki *et al.*, 1970; Montgomery *et al.*, 1972; Reynolds *et al.*, 1973).

Among the infants delivered by caesarean section intact fetal membranes did not prevent perinatal CMV infection. This differs from the situation in herpes simplex virus infections where caesarean section has been recommended as long as the membranes are intact in order to prevent the spread of the virus from the birth canal to the offspring. In the work of Reynolds *et al.* (1973) CMV could not be cultivated from the amniotic fluid of the mothers who excreted this virus in the cervix at the end of gestation. It is also noteworthy that the placentas of these infants were free from virus, and yet nearly 40% of the infants developed perinatal CMV infection. It was therefore strongly suspected that the principal route of infection is the birth canal (Reynolds *et al.*, 1973). *In vitro* studies have shown that CMV readily grows in endocervical epithelial cells of the uterine cervix (Vesterinen *et al.*, 1975). The endocervix might harbour CMV, and the fetus may possibly catch the virus from this location before birth. This assumption is corroborated by the occurrence of perinatal infection in infants delivered by caesarean section. The route of infection may be the same in asymptomatic congenitally infected infants.

Since CMV is excreted in maternal milk, this has been considered a possible route of transmission of the virus from mother to child (Hayes *et al.*, 1972). In the present series the perinatally infected infants were breast fed at the age of 2 months more often than the noninfected infants. However, there was no difference in the duration

of breast feeding between those who started excreting virus at the age of 2 or 4 months, respectively. Our results therefore do not eliminate the possibility that maternal milk may be the source of perinatal CMV infection.

The risk of acquiring perinatal CMV infection could not be related to maternal age, duration of pregnancy, or the relative weight of the placenta. The mothers of the congenitally infected infants were younger than the average at the time of delivery in the series of Starr *et al.* (1970). In our series 2 of the 3 mothers were under 22 years of age.

The only difference between perinatally infected and noninfected infants in Levinsohn's series (1969) was the clearly lower birthweight of the CMV infected infants. The mean birthweight in our series was the same in both groups and correlated well with the Finnish standards (Bäckström and Kantero, 1971). The birthweights of the congenitally infected infants were also in the normal range. However, among the infants with a birthweight under 3000 g the incidence of perinatal infection was higher than among those with a higher birthweight. Of 9 infants with a birthweight under 3000 g, a gestational age of over 37 weeks, and mothers with CMV antibodies, as many as 7 developed perinatal CMV infection. This differs clearly from the other birthweight groups and supports the opinion that risk of perinatal CMV infection is higher in small-for-dates babies.

Only one infant was mildly asphyxiated after delivery. The remainder had Apgar scores of at least 7 (Ikonen, 1973). One unexplained difference was observed: among the perinatally infected infants an Apgar score of 8 was more common than among the noninfected infants, who mostly received an Apgar score of 9.

The majority of the perinatally infected infants were born as the first child in the family. Comparing only the children of seropositive mothers, the risk for the first-born children of developing perinatal CMV infection was greater than the risk for the remainder. We do not know whether the older sibs of the perinatally infected infants had had CMV infections, but it may be stated that perinatal CMV infection is more prevalent in first-born children than among their sibs. This perhaps reflects a difference in the activation process of latent CMV infection during consecutive pregnancies.

Many authors have assumed that the white blood cells in fresh blood are the source of CMV infection in the post-transfusion syndrome (Lang and Han-shaw, 1969). Exchange transfusions have been suspected of transmitting virus to babies (Luthardt *et al.*, 1971; Yeager, 1974). 7 of our infants received fresh blood during the first week of life, and only 3 of them developed perinatal CMV infection. This

incidence is of the same order as in the total material. Exchange transfusion did not cause perinatal infection in the child of a seronegative mother.

An association between CMV infection during the first months of life and pulmonary or hepatic symptoms has been suggested (Stern, 1968), but we did not observe pneumonia or other respiratory illness or signs of hepatitis more often among the perinatally infected than among the noninfected infants. The perinatally infected infants did not have any features in common that could be considered symptoms or signs of CMV infection. 3 severe urinary tract infections caused by *E. coli* were seen in perinatally infected girls. Though the number is small, this might imply that a concomitant CMV infection of the urinary tract could aggravate the course of the bacterial infection.

The motor and psychosocial development was similar in the CMV infected and the noninfected infants. Neither congenital nor perinatal CMV infection increased the risk of CNS damage which could be observed during the first year of life in our patients. Of course slight developmental abnormalities cannot be recognized at this age but may be shown in later examinations of the groups. However, in several prospective studies the observed malfunctions of the central nervous system in congenitally infected infants have been obvious. Probably the number of congenitally infected infants in this study was too small to show symptomatic congenital infections. But the number of perinatally infected infants seems sufficient for the statement that the risk of unfavourable development caused by this type of CMV infection, if any, must be small.

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References

- Bäckström, L., and Kantero, R.-L. (1971). Cross-sectional studies of height and weight in Finnish children aged from birth to 20 years. *Acta Paediatrica Scandinavica*, **Suppl.** 220, 9-12.
- Hanshaw, J. B. (1971). Congenital cytomegalovirus infection: a fifteen year perspective. *Journal of Infectious Diseases*, **123**, 555-561.
- Hayes, K., Danks, D. M., Gibas, H., and Jack, I. (1972). Cytomegalovirus in human milk. *New England Journal of Medicine*, **287**, 177-179.
- Ikonen, R. S. (1973). Apgar scoring and neonatal morbidity in full-sized newborn infants. *Annals of Clinical Research*, **5**, 380-384.
- Kantero, R.-L., and Tiisala, R. (1971). Growth of head circumference from birth to 10 years. *Acta Paediatrica Scandinavica*, **Suppl.** 220, 27-32.
- Kibrick, S., and Loria, R. M. (1974). Rubella and cytomegalovirus. *Pediatric Clinics of North America*, **21**, 513-526.
- Lang, D. J., and Hanshaw, J. B. (1969). Cytomegalovirus infection and the postperfusion syndrome. Recognition of primary infections in four patients. *New England Journal of Medicine*, **280**, 1145-1149.
- Lapinleimu, K., Koskimies, O., Cantell, K., and Saxén, L. (1974). Association between maternal herpes virus infections and congenital malformations. *Lancet*, **1**, 1127-1129.
- Leinikki, P. (1972). Immunofluorescent antibody technique in herpes simplex virus type 2 infections. Thesis, Helsinki.
- Leinikki, P., Heinonen, K., and Pettay, O. (1972). Incidence of cytomegalovirus infections in early childhood. *Scandinavian Journal of Infectious Diseases*, **4**, 1-5.
- Levinsohn, E. M., Foy, H. M., Kenny, G. E., Wentworth, B. B., and Grayston, J. T. (1969). Isolation of cytomegalovirus from a cohort of 100 infants throughout the first year of life. *Proceedings of the Society for Experimental Biology*, **132**, 957-962.
- Luthardt, T., Siebert, H., Lösel, J., Quevedo, M., and Todt, R. (1971). Cytomegalievirusinfektionen bei Kindern mit Blutaustauschtransfusion im Neugeborenenalter. *Klinische Wochenschrift*, **49**, 81-86.
- Melish, M. E., and Hanshaw, J. B. (1973). Congenital cytomegalovirus infection. Developmental progress of infants detected by routine screening. *American Journal of Diseases of Children*, **126**, 190-194.
- Montgomery, R., Youngblood, L., and Medearis, D. N., Jr. (1972). Recovery of cytomegalovirus from the cervix in pregnancy. *Pediatrics*, **49**, 524-531.
- Numazaki, Y., Yano, N., Morizuka, T., Takai, S., and Ishida, N. (1970). Primary infection with human cytomegalovirus: virus isolation from healthy infants and pregnant women. *American Journal of Epidemiology*, **91**, 410-417.
- Reynolds, D. W., Stagno, S., Hosty, T., Tiller, M., and Alford, C. A., Jr. (1973). Maternal cytomegalovirus excretion and perinatal infection. *New England Journal of Medicine*, **289**, 1-5.
- Reynolds, D. W., Stagno, K., Stubbs, K. G., Dahle, A. J., Livingston, M. M., Saxon, S. S., and Alford, C. A. (1974). Inapparent congenital cytomegalovirus infection with elevated cord IgM levels. Causal relation with auditory and mental deficiency. *New England Journal of Medicine*, **290**, 291-296.
- Starr, J. G., Bart, R. D., Jr., and Gold, E. (1970). Inapparent congenital cytomegalovirus infection. Clinical and epidemiologic characteristics in early infancy. *New England Journal of Medicine*, **282**, 1075-1078.
- Stern, H. (1968). Isolation of cytomegalovirus and clinical manifestations of infection at different ages. *British Medical Journal*, **1**, 665-669.
- Stern, H., and Tucker, S. M. (1973). Prospective study of cytomegalovirus infection in pregnancy. *British Medical Journal*, **2**, 268-270.
- Stoll, H. U. (1974). Zytomegalievirusausscheidung bei gesunden Kindern im ersten Lebensjahr. *Schweizerische Medizinische Wochenschrift*, **104**, 468-472.
- Vesterinen, E., Leinikki, P., and Saksela, E. (1975). Cytopathogenicity of cytomegalovirus to human ecto- and endocervical epithelial cells in vitro. *Acta Cytologica*, **19**, 473-481.
- Yeager, A. S. (1974). Transfusion-acquired cytomegalovirus infection in newborn infants. *American Journal of Diseases of Children*, **128**, 478.

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