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Virologic and Serologic Studies on Infectious Mononucleosis-like Disease in Children, with Special Reference to Cytomegalovirus, Epstein-Barr Virus and Adenovirus Infections

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IKEDA, S., CHIBA, S., AGATSUMA, Y., SUZUKI, M., WATAYA, Y. and NAKAO, T. *Virologic and Serologic Studies on Infectious Mononucleosis-like Diseases in Children, with Special Reference to Cytomegalovirus, Epstein-Barr Virus and Adenovirus Infections.* Tohoku J. exp. Med., 1974, **112** (1), 47–61 —— The relationship between clinical manifestations and etiologic viruses was discussed based on 23 cases of Japanese children with infectious mononucleosis-like disease. 1) Five cases were caused by cytomegalovirus, 6 caused by Epstein-Barr virus, 2 caused by adenovirus, and 2 caused by a double infection with cytomegalovirus and Epstein-Barr virus. The other 8 cases were caused by unknown etiologic agents. 2) In the group due to cytomegalovirus the onset age was under 1 year, and the frequency of hepatosplenomegaly and liver involvement was high. In the group due to Epstein-Barr virus the onset age was over 1 year, and the frequency of pharyngitis and lymphadenopathy was high. 3) These characteristics of clinical manifestations of the diseases due to both viruses in Japanese children were almost coincident with those of infectious mononucleosis due to Epstein-Barr virus and cytomegalovirus mononucleosis seen in adults in America or Europe. ——— infectious mononucleosis-like disease; cytomegalovirus; Epstein-Barr virus; adenovirus

A viral etilogy for infectious mononucleosis (IM) has been suspected for many years. Recently, Epstein-Barr virus (EBV) found in cultured Burkitt lymphoma cells was reported to be related to the causative virus of IM by Henle et al.(19 68).

On the other hand Klemola and Kääriäinen (1965) reported that cytomegalovirus (CMV) could cause the disease hematologically resembling Paul-Bunnell test negative IM without pharyngitis and lymph nodes enlargement.

As the causative agents of IM-like disease adenovirus (ADV) (Gutekunst and Heggie 1961) and toxoplasma (Remington et al. 1962) have also been suspected. These both resembling diseases are most prevalent in adolescence in America and Europe, while reports about the diseases in children are scanty.

The purposes of this paper are to clarify 1) whether CMV, EBV and ADV are causative agents of infectious mononucleosis (IM)-like disease in Japanese children as in American and European adults or not, and 2) whether there is some

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TABLE 1. *Virologic and serologic data of 23 cases*

Case no.	Age	Sex	The results of virologic and	
			Isolation of CMV	Titers of CMV.CF antibody
1*	2yr. 1mo.	M	(8) urine (+) (8) saliva (+)	(8) 128 (32) ≧ 256
2*	5mo.	F	(10) urine (+) (10) saliva (+)	(10) 8 (30) 16 (120) 64
3	4yr. 6mo.	M	cont.	(9) 16 (22) 16 (33) 16
4*	5yr. 0mo.	M	(7) urine (−) (7) saliva (−)	(4) <4 (24) <4
5*	1yr. 0mo.	M	(10) urine (−) (10) saliva (−)	(9) <4 (30) <4
6	5yr. 7mo.	M	NT	(24) 32 (31) 32
7	3yr. 11mo.	M	NT	(9) <8 (19) <8
8	7yr. 7mo.	F	cont.	(7) 32 (30) 16
9	1yr. 1mo.	M	(9, 21) saliva (−)	(9) <8 (16) <8 (25) <8
10	8yr. 7mo.	F	NT	(7) <8 (14) <8 (20) <8
11	11yr. 10mo.	M	(22) urine (−)	(8) 16 (26) 16
12	6yr. 2mo.	F	(7) urine (−) Lymph node (−) WBC (−)	(4) <4 (24) <4 (60) <4
13	4mo.	M	(−14, 4) urine (+) saliva (+)	(3) 64 (14) ≧ 128 (23) ≧ 128
14	2mo.	F	(30) urine (+) (120) urine (+)	(30) ≧ 64 (120) 32 (270) 16
15	5mo.	M	cont.	(7) 8 (24) 16 (34) 32
16	1yr. 8mo.	F	cont.	(24) 32

with infectious mononucleosis-like disease

serologic studies on IM-like disease			
Titers of EBV antibody		Titers of ADV.CF antibody	Causative virus
Anti-V	Anti-N		
NT	NT	8	
		8	CMV
20	<10	16	
40	<10	8	CMV
40	<10	4	
160	20	<8	
160	20	<8	EBV
160	20	<8	
NT	NT	16	unknown
		16	
		<4	ADV
10	<10	16	
160	<10	<8	EBV?
160	<10	<8	
40	<10	8	unknown
40	<10	8	
		<8	unknown
80	<10	<8	
640	20	<8	EBV
640	10	<8	
20	<10	<8	
			unknown
40	<10	<8	
10	<10	<8	EBV
40	<10	<8	
<10	<10	8	
<10	<10		unknown
<10	<10	8	
80	<10	<8	
80	<10		CMV
80	<10	<8	
20	<10	<8	
20	<10	<8	CMV?
10	<10	<8	
10	<10	<8	CMV
20	<10		and/or
80	<10	<8	EBV
160	40	<8	EBV

TABLE 1.

Case no.	Age	Sex	The results of virologic and	
			Isolation of CMV	Titers of CMV.CF antibody
17	5yr. 10mo.	M	(29) urine (−)	(20) <4 (180) <4
18	2yr. 11mo.	F	(7) saliva (−)	(5) ≧ 64 (23) ≧ 64 (34) ≧ 64
19	1yr. 1mo.	M	NT	(34) <8
20	4mo.	F	(26 after admission) urine (−)	after admission (5) <8 (30) <8 (60) <8
21	1yr. 0mo.	M	(19, 48) urine (−) saliva (−)	(17) 32 (48) 32 (120) 32
22	2mo.	M	(19) urine (+) (19) saliva (+)	(5) 32 (19) ≧ 128
23	2yr. 4mo.	M	NT	(16) ≧ 128 (60) 16

* Previously reported cases (Chiba et al. 1969) ( ), days after onset

relationship between causative viruses and clinical manifestations of IM-like disease or not.

MATERIALS AND METHODS

Twenty-three children with IM-like disease were admitted to the hospital of Sapporo Medical College, Sapporo, from January 1966 to July 1972. Diagnostic criteria of IM-like disease were mainly based on clinical and hematologic findings. Some of these cases were reported previously (Nakao et al. 1967, Chiba et al. 1969).

The serial serum samples obtained during the course of the disease were stored at −20°C until simultaneously tested.

Antibodies to CMV and ADV were assayed by the complement-fixation (CF) test. CF antigen of CMV was prepared from the strain Ad 169 in human embryonic fibroblasts. The technical details of the procedures were described previously (Chiba et al. 1968). CF antigen of ADV (type 3) was obtained commercially (Toshiba Chemical Laboratories, Tokyo). The CF test was carried out by the microtechnique using 4 units of antigen (Sever 1962). Four-fold rise or decline in antibody titer was considered to be significant.

Antibodies to virion (V) antigen and nonstructual (N) antigen of EBV were detected by the indirect-immunofluorescence method. The indirect-immunofluorescence test for EBV was carried out at the Department of Microbiology, Kumamoto University Medical School, Kumamoto. The method used is described in detail elsewhere (Hinuma et al. 1967, Ida et al. 1972).

Some of the patients were examined to isolate CMV from their urine and saliva specimens using human embryonic fibroblasts. Details of isolation and identification of CMV are also previously described (Chiba et al. 1968).

Continued

serologic studies on IM-like disease			
Titers of EBV antibody		Titers of ADV.CF antibody	Causative virus
Anti-V	Anti-N		
80	<10	32	
80	<10	<8	ADV
20	<10	16	
20	<10		unknown
20	<10	16	
<10	<10	8	unknown
20	<10	<8	
20	<10	<8	unknown
20	<10	<8	
<10	<10	8	
40	10	8	EBV
160	20	8	
20	<10	<8	
20	<10	<8	CMV
640	<10	16	CMV and/or
640	10	8	EBV

NT, not tested cont., contamination (+), positive (−), negative

Results

The results of virologic and serologic studies of IM-like disease are shown in Table 1. Five cases were caused by CMV, and 6 cases caused by EBV. Two cases were suspected to be caused by CMV and/or EBV. Two cases were caused by ADV, and the other 8 cases caused by unknown agents. Among the group due to unknown agents Case 12, a 6-year and 2 month-old girl, had a high level of ASO units, and Case 20, a 4-month-old female, had ever been received a blood transfusion for melena neonatorum.

Table 2 shows results of hematologic findings and serologic reactions of IM-like disease. In 3 cases (Cases 9, 14 and 19) Paul-Bunnell test were positive, but Davidsohn test gave negative results.

The relationship between clinical manifestations and causative viruses is shown in Table 3.

Four infants were under 1 year of age out of 5 cases due to CMV. On the other hand all the 6 cases due to EBV were over 1 year of age.

Only 1 case suffered from pharyngitis among the group due to CMV. Five cases suffered from pharyngitis among the group due to EBV.

Lymphadenopathy was found in only 1 case among the group due to CMV, but in 4 cases among the group due to EBV.

TABLE 2. *Hematologic findings and serologic reactions of 23 cases with infectious mononucleosis-like disease*

Causative virus	Case no.	Age	Sex	White blood cells			Serologic reactions	
				Total ($\times 10^3$)	Atypical lympho-cytes (%)	Lympho-cytes (%)	Paul-Bunnell test	ASO (Todd units)
Group due to CMV	1*	2yr. 1mo.	M	18.6	18.0	33.5	112 112 56	NT
	2*	5mo.	F	32.4	15.0	46.0	7 7	NT
	13	4mo.	M	3.0	23.0	69.0	28	NT
	14	2mo.	F	17.6	13.0	63.5	224 112	NT
	22	2mo.	M	14.4	14.5	59.0	NT	NT
Group due to EBV	3	4yr. 6mo.	M	19.6	25.5	55.0	112 112 112	NT
	6	5yr. 7mo.	M	9.9	15.5	27.0	56 56 56	833 250
	9	1yr. 1mo.	M	15.6	17.0	24.5	224 448 224	<12
	11	11yr. 10mo.	M	5.5	3.5	68.0	128 112 56	NT
	16	1yr. 8mo.	F	19.4	21.5	55.5	112 56	<12
	21	1yr. 0mo.	M	11.5	39.0	38.0	28 14	NT
Group due to ADV	5*	1yr. 0mo.	M	15.0	15.5	24.0	7 14 14	<12
	17	5yr. 10mo.	M	7.6	12.5	58.0	56 28	<12
Group due to CMV and/or EBV	15	5mo.	M	24.0	13.5	54.5	28 56	<12
	23	2yr. 4mo.	M	5.4	58.0	14.0	112 56	<12
Group caused by unknown agents	4*	5yr. 0mo.	M	17.2	26.6	22.0	14 7 28	12

TABLE 2. *Continued*

Causative virus	Case no.	Age	Sex	White blood cells			Serologic reactions	
				Total ($\times 10^3$)	Atypical lympho-cytes (%)	Lympho-cytes (%)	Paul-Bunnell test	ASO (Todd units)
Group caused by unknown agents	7	3yr. 11mo.	M	18.8	40.0	40.0	112 14 56	166
	8	7yr. 7mo.	F	11.4	55.0	25.0	14 28	166
	10	8yr. 7mo.	F	20.6	13.5	37.0	56 56	500
	12	6yr. 2mo.	F	14.1	30.0	31.0	56 112 28	>2500 625
	18	2yr. 11mo.	F	11.8	7.5	74.0	14 28	50 50
	19	1yr. 1mo.	M	10.4	16.5	24.5	1792 1792 1792	<12
	20	4mo.	F	11.3	7.5	74.0	14 28	50 50

* Previously reported cases (Chiba *et al.* 1969) NT, not tested

Hepatomegaly and splenomegaly were found in 5 and 4 cases among the group due to CMV, respectively. Among the group due to EBV both manifestations were found in only 1 case respectively.

A measles-like rash appeared at the trunks and extremities in 1 case due to CMV, EBV and ADV, respectively.

The relationship between routine laboratory findings and causative viruses is shown in Table 4. In 4 cases among the group due to CMV liver function tests (SGOT and SGPT) gave abnormal results. On the other hand only 2 cases displayed liver impairment among the group due to EBV. But none of the patients with liver impairment were icteric.

Only 1 case among the group due to CMV showed the enlargement of hilar marking in the chest x-ray film. Four cases showed the enlargement of hilar marking among the group due to EBV.

The relationship between serologic results and causative viruses is shown in Table 5.

Two cases displayed Paul-Bunnell titer in 1:112 or greater among the group due to CMV, on the other hand 4 cases displayed the titer in 1:112 or greater among the group due to EBV. But in all the positive cases Davidsohn tests gave negative results.

The prognosis of all the cases was good.

TABLE 3. *Relationship between clinical manifestations and*

Causative viruses	No. of cases over 1 year of age	Clinical	
		Highest fever (≧38°C)	Pharyngitis
CMV Five cases	1/5 (20)	4/5 (80)	1/5 (20)
EBV Six cases	6/6 (100)	3/6 (50)	5/6 (83)
ADV Two cses	2/2	2/2	1/2
CMV and/or EBV Two cases	1/2	2/2	1/2
Unknown Eight cases	7/8	7/8	7/8

( ), percentage

TABLE 4. *Relationship between routine laboratory findings and causative viruses in 23 cases with infectious mononucleosis-like disease*

Causative viruses	Routine laboratory findings of IM-like disease		
	ESR (≧20 mm/hour)	Increase in SGOT and SGPT (maximal value according to Reitman & Frankel)	Enlargement of hilar markings in chest X-ray film
CMV Five cases	2/4 (50)	4/5 (80)	1/4 (25)
EBV Six cases	3/6 (50)	2/6 (33)	4/6 (67)
ADV Two cases	1/2	1/2	1/2
CMV and/or EBV Two cases	2/2	1/2	1/2
Unknown Eight cases	7/8	7/8	5/8

( ), percentage

CASE REPORT

The clinical courses of the 4 representative cases whose causative viruses were distinctive were described below.

Y.K. (Case 9), a 1-year and 1 month-old male infant, had cough, stridor and high fever since July 22, 1969. A measles-like rash appeared on his face on July 30, and spread to his trunk. He was admitted to our clinic with the eruption and pharyngitis on August 1, 1969. On admission cervical lymph nodes were

causative viruses in 23 cases with infectious mononucleosis-like disease

manifestations of IM-like disease			
Lymphadenopathy	Hepatomegaly	Splenomegaly	Exanthema
1/5 (20)	5/5 (100)	4/5 (80)	1/5 (20)
4/6 (67)	1/6 (17)	1/6 (17)	1/6 (17)
2/2	1/2	1/2	1/2
1/2	1/2	0/2	0/2
7/8	4/8	2/8	2/8

TABLE 5. *Relationship between serologic findings and causative viruses in 23 cases with infectious mononucleosis-like disease*

Causative viruses	Serologic results	
	Paul-Bunnell test (≧112)	ASO (≧333 Todd u.)
CMV Five cases	2/5 (40)	NT
EBV Six cases	4/6 (67)	2/4 (50)
ADV Two cases	0/2	0/2
CMV and/or EBV Two cases	1/2	0/2
Unknown Eight cases	4/8	2/7

( ), percentage NT, not tested

palpable. Hepatosplenomegaly was not recognized. Atypical lymphocytes occupied 17% of peripheral white cell counts. Paul-Bunnell test was positive (1:224), but Davidsohn test was negative (<1:14). He was discharged on September 2, 1969. At the time of his discharge cervical lymph nodes were not palpable, atypical lymphocytes counts 0.5% and erythrocyte sedimentation rate 7 mm/hr. Our attempts failed to isolate CMV from his urine and saliva. The titers of CF antibodies against CMV and ADV were negative in this case. The titer of fluorescent antibodies against V-antigen of EBV was 1:640 (on 9 and 25 days after onset of the disease), and the titer against N-antigen was 1:20 (on 9 days after onset)

and 1:10 (on 25 days after onset). The causative virus was suspected to be EBV in this case.

T.S. (Case 17), a 5-year and 10 month-old boy, had a high fever of 39.5°C and was treated as pharyngitis since June 1, 1971. He had a maculopapular measles-like rash over his trunk and extremities, and palpable lymph nodes in his neck, axillae and groins. At that time hepatosplenomegaly was noticed. He was admitted to our clinic with 12% atypical lymphocytes on July 7, 1971. Thirteen days after admission he was discharged with only mild abnormal values in the liver function. He had a slight enlargement of cervical lymph nodes, and the liver was palpable at 1.5 cm below the right costal margin at the midclavicular line at his discharge. The titers of CF antibodies against CMV and ADV or fluorescent antibody titers against EBV were determined in the sera obtained during his admission and 2 month after his discharge. Only the CF antibody titers against ADV responded significantly in the paired sera (titer 1:32 on 20 days after onset; <1:8 on about 90 days after onset). Cultures for CMV were negative from his urine. ADV infection was suspected in this case.

K.Y. (Case 21), a 1-year-old male infant, had rhinorrhea and cough, and his face became pale on February 21, 1972. He was admitted to our clinic with a fever of 38.2°C, enlargement of cervical lymph nodes and hepatosplenomegaly. There was a change in the white blood cells distribution to a predominance of mononuclear cells with 39.0% atypical lymphocytes. Isolations of CMV from his urine and saliva gave negative results. Antibody titers were determined against CMV, ADV and EBV in the serially obtained sera during the disease.

Fluorescent antibody titers against V- and N-antigen of EBV rised significantly (anti-V <1:10 and anti-N <1:10 on day 17; anti-V 1:40 and anti-N 1:10 on 48 days after onset; anti-V 1:60 and anti-N 1:20 on about 90 days after onset), and EBV infection was suspected in this case.

S.T. (Case 22), a 2-month-old male infant, was noticed hepatosplenomegaly in the Health Center in Sapporo City and admitted to our hospital for detailed examination on April 15, 1972.

On admission the liver was palpable 3 cm below the right costal margin at the midclavicular line, and the spleen 5 cm, and cervical lymph nodes were palpable. He had no fever and his general condition was good.

Mononuclear cells dominated the differential count with 14.5% atypical lymphocytes. Moderate damage of liver function was recognized.

Successful isolations were made from his urine and saliva in this case. Antibody titers were determined against CMV, ADV and EBV in the serially obtained sera during his admission. CF antibody titers against CMV rised significantly in the paired sera (titer 1:32 on 15 days after onset; 1:128 on 19 days after onset). Diagnosis of CMV infection was established in this case.

Discussion

Until recently the causative agent of IM has been unknown, although a viral etiology has been postulated for a long time.

Henle and his associates (Evans et al. 1968, Henle et al. 1968, Niederman et al. 1968) showed that many of the cases with typical IM developed fluorescent antibodies against EBV found in cultured Burkitt lymphoma cells. Thereafter many investigators (Gsell et al. 1970, University Health Physicians and P.H.L.S. Laboratories 1971, Goetz and Peller 1971) reported the same results. It is a general belief to recognize that EBV plays a most important role in the causative agents of IM.

According to the reports by Niederman et al. (1968) clinical manifestations of IM due to EBV were as follows; protracted fever, pharyngitis, lymphadenopathy, appearance of atypical lymphocytes and positive Paul-Bunnell test.

On the other hand, Klemola and his associates (Klemola and Kääriäinen 1965, Kääriäinen et al. 1966) reported that CMV caused a disease resembling IM. The disease was characterized by protracted fever, liver involvement, absence of pharyngitis and lymph nodes enlargement, atypical lymphocytes in peripheral blood, and negative results on Paul-Bunnell test. The name cytomegalovirus mononucleosis has been proposed for this disease by them.

According to our observations the frequency of pharyngitis and lymph node enlargement was high in the group due to EBV compared with the group due to CMV. Paul-Bunnell test positive cases were more frequent in the group due to EBV. To the contrary, Paul-Bunnell test negative cases were more frequent in the group due to CMV, and the frequency of hepatosplenomegaly and liver involvement was high compared with the group due to EBV. These characteristics in clinical findings of the both diseases were almost coincident with those of IM or IM-like disease seen in American and European adults.

The antibody response in CMV or EBV infection differs somewhat from the one of the other viral infection. In a majority of patients EBV antibodies tended to appear early and reached peak levels within 3 to 4 weeks after the onset of symptoms (Niederman et al. 1968). But in some patients EBV or CMV antibodies were considerably delayed in their appearance. For example EBV antibodies did not reach significant levels until the 3rd week of illness in 2 cases out of 27 cases observed by Niederman et al. (1968). According to the report of Klemola et al. (1970) CF antibody titers against CMV reached peak levels between 4 and 7 months after onset of the symptoms of CMV mononucleosis. As shown in our previous report (Chiba et al. 1969) in some patients with IM-like disease CF antibody titers against CMV developed gradually and reached peak levels in 1:256 7 months after onset of the disease.

In this series Case 21, a 1-year-old male infant, showed the delayed fluorescent antibody response against V- and N-antigen of EBV, and the titers reached peak levels 3 months after the onset of the disease.

N-antigen of EBV found by Hinuma et al. (1970) was similar to the early

antigen (EA) of EBV found by Henle et al. (1970). Henle et al. (1971) said that since the duration of anti-EA response was transitory, the mere presence of the antibody in a serum indicated a current or recent primary EBV infection. They said also that in Paul-Bunnell negative IM-like disease the detection of anti-EA strongly supported this diagnosis. Henle et al. (1971) observed that anti-EA was found in sera from patients with IM but not in those from healthy donors. In contrast, Ida et al. (1972) said that this similar antibody (anti-N) was not detected in sera from 19 patients with IM. They suspected that the cases of IM in Japan might not be caused by EBV but by other etiologic agents.

According to our observations some of the patients with IM-like disease had the significant rises of anti-N.

Hinuma et al. (1969) examined the age distribution of EBV antibodies among Japanese, and it was found that the incidence of the positive sera was extremely high in infancy and the level remained high in adult age. They revealed that the incidence of individuals with antibody reached about 80% by 3 years of age. The results strongly suggested that EBV infection was established during infantile period in most Japanese. In contrast, Henle and Henle (1967) reported that the age distribution of positive sera increased to about 50% by 4 years of age.

In America or Europe IM or IM-like disease occurred mostly in adolescence. In contrast, these diseases were most prevalent in childhood in Japan, and in a majority of the cases Paul-Bunnell test were negative. The high incidence of EBV infection during infancy may suggest a possible vertical transmission, probably in utero infection, in most Japanese (Hinuma et al. 1969).

The reasons still remain unknown why the Paul-Bunnell test negative IM is popular in children. The reasons are suspected as follows; there may be a specificity of the reaction on the Paul-Bunnell test in the sera of children, or a majority of IM in Japan may not be caused by EBV but by other etiologic agents.

According to our observations the mean age of the patients due to EBV was over 1 year. In contrast, the mean age of the patients due to CMV was under 1 year.

Evans et al. (1968) stated that EBV antibody was also present in the sera of the patients who had negative results on the Paul-Bunnell test as the patients with positive results. These findings suggested that EBV might be associated with both forms of illness. They said also that among children Paul-Bunnell test negative and EBV antibody positive cases appeared to be more common.

Hoagland (1955) stated that the etiologic agent of IM was transmitted chiefly by the direct intimate oral contact which allowed for salivary exchange, and stated also that the incubation period of the disease was about 33 to 49 days. Ödegaard (1967) investigated 911 cases of IM and suspected that the agent would be transmitted by salivary exchange through kissing. Heath et al. (1972) reported that the contact was recognized in 11 cases with IM of the opposite sex. In their observation the time interval between contact and diagnosis ranged from 0 to 7 weeks with a mean of 38 days.

Golden et al. (1971) reported that throat washings from 6 out of 7 patients with IM contained filtrable agent which converted an indicator lymphocyte from negative to positive for EBV antibodies by the indirect immunofluorescence test. Pereira et al. (1972) found herpes virus particles in the transformed lymphocytes with throat swab material taken from 1 case out of 6 cases with IM by electron microscopy. They also suspected this herpes virus as EBV by immunofluorescence. Miller et al. (1973) found a factor that transformed human and simian blood leukocytes into continuous cell lines in throat washes from 23 of 25 patients with IM. They suspected that the transforming factor was EBV. These findings show that direct oral contact will play a most important role in infective routes of IM.

Wising (1942) transferred into a volunteer blood from a patient with IM. Twelve days after transfusion atypical lymphocytes appeared and on the 16th day fever, sore throat and malaise occurred. On the 24th day heterophile antibodies which had been negative before the transfusion developed to 1:32. EBV was detected in all cultures which were initiated with peripheral leucocytes from the patients with IM by immunofluorescence and also by electron microscopy (Diehl et al. 1968).

Kääriäinen et al. (1966) stated that CMV infection would occur by the transfusion of fresh blood during open heart surgery. Foster and Jack (1968) isolated CMV from peripheral lymphocytes of the patient with post-transfusion mononucleosis.

Wahren et al. (1969) reported two out of 11 with CMV antibody rises were also found to have antibody rises to EBV. The possibility of double infections with CMV and EBV was discussed by Evans (1972). In our observations 2 cases were suspected to be simultaneously infected with EBV and CMV.

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