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APPLICATION OF A LIVE ATTENUATED VARICELLA VACCINE TO HOSPITALIZED CHILDREN AND ITS PROTECTIVE EFFECT ON SPREAD OF VARICELLA INFECTION

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SUMMARY Twenty-three hospitalized children with no history of varicella or no detectable complement fixing (CF) antibody, were vaccinated with a live attenuated varicella vaccine (Oka strain) immediately after the occurrence of a case of varicella in a children's ward of hospital. These children suffered from the nephrotic syndrome, nephritis, purulent meningitis, hepatitis etc., and 12 of them were receiving steroid therapy. An antibody response was noticed in all the vaccinated children, with mild fever in 6 and a mild rash in 2 of the 6. It was uncertain whether these reactions were due to vaccination or to naturally acquired infection modified by vaccination. No other clinical reactions or abnormalities of the blood or urine were detected. Thus the spread of varicella infection was prevented, with the exception of one severe case in an unvaccinated patient.

In another trial, 16 children with renal diseases were also vaccinated. All the children showed an immune response with no clinical reactions and no abnormalities in blood and urine examinations.

Thus live varicella vaccine (Oka strain) can be used safely and effectively for hospitalized children, and its effectiveness in preventing spread of varicella infection was confirmed.

INTRODUCTION

Varicella is a relatively mild illness, which is commonest in childhood and rarely causes complications in normal children. However, when it occurs in the children's ward of a hospital, it is well known that it may be serious and sometimes lethal in patients with leukemia or other malignancies (Cheatham et al., 1956), as well as in patients receiving im-

munosuppressive treatments (Haggerty and Eley, 1965; Gershon et al., 1972) and those with congenital immunodeficiencies (Johnston and Janeway, 1969). Recently, the increasing risk of varicella infection has been recognized in many hospitals, since immunosuppressive drugs are now universally used for treatment of diseases such as the nephrotic syndrome. It has been observed that simple quarantine usually fails to prevent nosocomial spread of varicella among sick children, so numerous attempts have been made to prevent or to modify the disease by passive immunization with convalescent serum (Weech, 1924), gamma-globulin (γ-G) (Funkhouser, 1948; Schaeffer and Toomey, 1948; Ross, 1962) or zoster immune globulin (ZIG) (Brunell et al., 1969, 1972, 1973; Gershon et al., 1974). Cytosine arabinoside (ARA-C) has also been used for treatment of disseminated zoster and varicella in compromised patients (Juel-Jensen, 1970; Prager, 1971; Stevens and Merigan, 1972; Stevens et al., 1973). But the effectiveness of these treatments varies, and in some instances clinical use of these agents may cause problems.

Moreover there has been no remarkable progress in developing a vaccine for varicella for active immunization. Recently, Takahashi et al. developed a live attenuated vaccine which induced an antibody response without clinical reactions in normal susceptible children (Takahashi et al., 1975). We employed this vaccine in a closed population of hospitalized children immediately after exposure to varicella. We found that the vaccine prevented nosocomial spread of varicella in the children's ward and that the vaccinees responded serologically without showing adverse clinical reactions, though they were with diseases such as the nephrotic syndrome, and some of them were receiving immunosuppressive therapy. Based on these findings, a further immunization trial was carried out on other patients of our renal clinic. The present paper reports details of these trials.

MATERIALS AND METHODS

1. *Vaccine*

The live attenuated varicella vaccine used was the Oka strain of varicella virus which had been passaged in human embryo lung (HEL) cells 11 times and then passaged in guinea pig embryo (GPE) cells 6 or 7 times. The preparations contained $10^{2.75}$ $TCID_{50}$/0.1 ml and $10^{2.5}$ $TCID_{50}$/0.1 ml, respectively (Takahashi et al., 1975). Virus was stored at -80 C until use.

2. *Vaccination*

Sick children who had no history of varicella were inoculated subcutaneously with 0.5 ml/dose of vaccine. Clinical observations were made daily for 10 weeks after vaccination. Blood samples were taken at intervals of one to two weeks for hematological, chemical and serological examination. These included examination of blood urea nitrogen, plasma total cholesterol, and serum creatinine, total protein, albumin, globulin, transaminase (SGOT, SGPT), sodium, potassium, chloride and immunoglobulins (IgG, IgA and IgM). Urine examinations were carried out every two days.

3. *Complement fixation tests*

CF antigen of varicella virus was prepared from HEL cell cultures infected with the Kawaguchi strain of varicella virus (Takahashi et al., 1975). Titration of CF antibodies was carried out in the Clinical Laboratory of the Chukyo Hospital by the standard microtiter technique described by Sever (1962), using 4 units of antigen, 2 units of complement and overnight fixation at 4 C. Uninfected cell antigen was also used at the same dilution of 4 units of antigen as a control.

RESULTS

1. *Vaccination of hospitalized children immediately after the occurrence of a case of varicella*

A 3 year old nephrotic boy developed clinical symptoms of varicella at the 1st Children's Ward of the Chukyo Hospital. Later the diagnosis was confirmed serologically. At the time of occurrence of varicella, there were 24 children in the Ward, 10 with a history of varicella. In the 2nd Children's Ward,

TABLE 1. *Clinical and serological responses in hospitalized children given a live varicella vaccine immediately after the occurrence of a case of varicella (Chukyo Hospital)*

Patient	Age (years)	Sex	Underlying disease	Steroid therapy	CF antibody titer					Fever			Rash	Laboratory findings (urine, blood)
					Pre	1 wk	2 wk	4 wk	10 wk	Onset (day)	Max temp (C)	Duration (days)		
S. H.[c]	4	F	Purpura	+	<4	—	32	64	16	13	37.7	1	—	—
K. O.	3	F	Myelitis	+	<4	—	32	32	—		—		—	—
U. K.	6m[e]	M	Hepatitis	+	<4	—	—	32	—		—		—	—
S. M.	1	M	Enteritis	—	<4	—	—	32	—		—		—	—
T. M.	8	M	Arthritis	+	<4	—	32	32	8		—		—	—
T. N.	11	M	Nephritis	—	<4	—	—	16	—		—		—	—
A. M.	4	F	Asthma	—	<4	—	16	32	—		—		—	—
K. S.	4	M	Enteritis	—	<4	—	4	32	16	10	37.5	1	—	—
T. S.	1	M	Hepatitis	+	<4	—	16	128	32	14	37.5	1	+	—
M. K.	1	M	Pur. mening.[a]	+	<4	—	16	16	—	14	37.5	1	—	—
S. N.	1	M	Pur. mening.	+	<4	—	—	8	—		—		—	—
M. Y.	1	F	Hemangioma	+	<4	—	—	32	—		—		—	—
M. H.	1	F	V.S.D.[b]	—	<4	—	64	64	32		—		—	—
T. S.	1	M	Hepatitis	+	<4	—	—	16	—		—		—	—
Y. T[d]	12	M	Nephrosis	+	<4	—	—	32	—		—		—	—
Y. I.	4	M	Nephrosis	+	<4	<4	16	64	32		—		—	—
A. K.	5	F	Nephritis	+	<4	<4	4	32	—	11	37.5	1	—	—
Y. I.	5	F	Nephritis	—	<4	<4	—	32	—		—		—	—
A. N.	1	F	Nephritis	—	<4	—	32	16	—	11	38.2	1.5	+	—
A. S.	3	F	Nephritis	—	<4	4	32	—	—		—		—	—
Y. M.	1	M	Nephritis	—	<4	—	—	16	—		—		—	—
M. A.	1	F	Enteritis	—	<4	—	—	32	—		—		—	—
K. N.	3	M	Nephritis	—	<4	<4	—	32	—		—		—	—

[a] Purulent meningitis. [b] Ventricular septal defect. [c] 1st Children's Ward.
[d] 2nd Children's Ward. [e] 6 months.

there were 30 children, 19 with a history of varicella. Therefore, to minimize spread of varicella infection, the varicella patient was transferred to the 2nd Ward where more children had a history of varicella. Then the 25 children with no history of varicella were immediately inoculated with varicella vaccine (Oka strain, HEL 11 times and then GPE 6 times passaged). CF antibody was not detectable in 23 of 25 children at a serum dilution of 1:4. These children suffered from the nephrotic syndrome, nephritis, purulent meningitis, asthma, hepatitis etc. and 12 of the 23 seronegative patients were receiving adrenocortical steroid hormone.

All the vaccinees became seropositive, and the mean CF antibody titers 4 weeks after vaccination were $2^{4.8}$ and $2^{5.0}$ in children not treated and treated with steroid, respectively, the difference not being significant.

None of the vaccinees showed any abnormal reactions in hematological, serum and urinary examinations during 10 weeks after vaccination. Six of the 23 vaccinated children developed mild fever of 37.5 C to 38.2 C, for 1 to 1.5 days 10 to 14 days after vaccination.

TABLE 2. *Clinical and serological responses in children with renal diseases given a live varicella vaccine (Chukyo Hospital)*

Patient	Age (years)	Sex	Underlying disease	Steroid therapy	CF antibody titer			Fever	Rash	Laboratory findings (urine, blood)
					0 wk	2 wk	4 wk			
YK	6	M	NS[a]	+	<4	—	128	—	—	—
TN	4	M	NS	+	<4	—	16	—	—	—
YK	1	F	NS	+	<4	—	32	—	—	—
GK	3	M	NS	−	<4	—	16	—	—	—
TK	4	M	NS	−	<4	—	16	—	—	—
KS	10 m[d]	M	N[b]	−	<4	16	—	—	—	—
KS	3	F	N	−	<4	16	—	—	—	—
TK	5 m[e]	M	N	−	<4	—	16	—	—	—
KH	6	F	N	−	<4	—	16	—	—	—
NI	1	M	N	−	<4	—	16 (8 wk)	—	—	—
TT	6	M	AGN[c]	−	<4	—	16	—	—	—
DY	4	M	AGN	−	<4	<4	8	—	—	—
MK	10	M	N	−	<4	—	8	—	—	—
AF	7	F	AGN	−	<4	—	32	—	—	—
KO	4	F	AGN	−	<4	—	64	—	—	—
IK	1	F	N	−	<4	—	32	—	—	—

[a] Nephrotic syndrome. [b] Nephritis (prolonged). [c] Acute glomerulonephritis.
[d] 10 months. [e] 5 months.

In 2 of them, a mild vesicular rash appeared. It was impossible to determine whether these reactions were due to the vaccine itself or to naturally acquired infection modified by vaccination. Except for these reactions, no clinical symptoms were detected and there was no spread of varicella among the hospitalized children.

In contrast, a 11-year-old nephrotic boy in the 2nd Children's Ward, who had not been vaccinated because his mother mistakenly believed that he had a history of varicella, developed severe symptoms of varicella 2 weeks after introduction of the 1st varicella patient into the 2nd Children's Ward. Thus it is evident that vaccination was effective in preventing spread of varicella among the hospitalized children, resulting in active immunity.

2. *Vaccination of children with renal diseases*

In another trial, 16 children with nephritis or the nephrotic syndrome, most of whom were hospitalized and some of whom were receiving steroid therapy, were inoculated with the vaccine (Oka strain, passaged 11 times in HEL and then 7 times in GPE). These children had no history of varicella or detectable CF antibody before vaccination. After vaccination all the children exhibited an antibody response without clinical reactions, and there were no unusual laboratory findings in blood or urine examinations during 4 weeks observation period.

DISCUSSION

Varicella virus infection in children receiving immunosuppressive thereapy for treatment of diseases such as the nephrotic syndrome can be serious or even fatal (Finkel, 1961; Grunberg, 1968; Sheinman, 1969). Complications of varicella can also be severe in other compromised patients, such as those with malignant diseases (Cheatham et al., 1956; Finkel, 1961),

immunosuppression (Haggerty and Eley, 1965; Gershon et al., 1972), or congenital deficiencies of the immune system (Johnston and Janeway, 1969). Therefore, many trials have been made to prevent varicella by passive immunization with γ-G, or with ZIG (Funkhouser, 1948; Schaeffer and Toomey, 1948; Ross, 1962; Brunell et al., 1969, 1972, 1973; Gershon et al., 1974), or to treat severe varicella and zoster with ARA-C (Juel-Jensen, 1970; Prager et al., 1971; Stevens and Merigan, 1972; Stevens et al., 1973). Ross (1962) demonstrated that the clinical course of varicella could be modified by giving γ-G to exposed, susceptible children within 72 hr after close contact with the infection, but varicella could not be prevented in this way. Brunell (1969) reported that ZIG extracted from plasma containing high titers of V-Z CF antibody was effective in preventing varicella if a 2 ml dose was given to susceptible children within 72 hr after household exposure. But immunocompromised children treated with ZIG were not protected against varicella as successfully as normal children (Brunell et al., 1972; Gershon et al., 1974). Brunell reported that five of nine high risk children who received ZIG within 48 hr after household exposure did not develop varicella, but three of the nine developed mild varicella with prolongation of the incubation period and one of the nine developed severe varicella. Recently, Gershon (1974) reported prophylactic efficacy of ZIG. In his report, 15 seronegative children received 3 to 5 ml of ZIG within 72 hr of household exposure to varicella and subsequently, varicella was severe in one, mild in nine and subclinical in five of the children. These observations indicate that ZIG can modify the clinical course of varicella in immunocompromised children with a more prolonged incubation period, milder clinical appearance and increase in the rate of subclinical infection. However, its effect is incomplete and it is not easy to obtain ZIG routinely. Moreover, there are many problems in treatment of severe cases of zoster or varicella with ARA-C because of its pharmaceutical effect (Juel-Jensen, 1970; Prager et al., 1971; Stevens and Merigan, 1972; Stevens et al., 1973).

Recently a live attenuated varicella vaccine has been developed which induces an antibody response without clinical reactions in normal children (Takahashi et al., 1975). We applied this vaccine to the hospitalized children in an attempt to prevent spread of varicella among sick children with no history of varicella when one of the hospitalized children exhibited the typical clinical symptoms of varicella. Although about half the vaccinated children were receiving immunosuppressive therapy, after vaccination no troublesome clinical reactions or disturbances of the blood or urine were detected. In contrast, one hospitalized child, who unfortunately was not vaccinated because of his mother's mistaken report, developed severe symptoms of varicella. Thus it is evident that the vaccination was effective in preventing spread of varicella among hospitalized children with no history of varicella. An antibody response was detected in all the vaccinated children and no difference was noticed in the mean antibody titer of children who were treated with steroid and those who were not treated. CF antibody could be detected as early as 2 week after vaccination.

Based on these results, the vaccine was applied to 16 children with nephritis or the nephrotic syndrome, some of whom were receiving steroid therapy. An antibody response was noticed in all the vaccinated children and no clinical reactions due to vaccination were observed. Diseases such as the nephrotic syndrome or nephritis are usually aggravated in naturally acquired varicella infection, but throughout our study no disturbances were observed in hematological, blood chemical or urinary examinations of any of the vaccinees.

More information is required on the persistency of the virus in the human body and the possible relationship between varicella virus and oncogenicity. However, at present, active immunization with live attenuated vac-

cine seems to be the most appropriate method to prevent natural infection of varicella in children who have underlying diseases such the nephrotic syndrome or nephritis.

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