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# Immunogenicity and Adverse Effects of Live Attenuated Varicella Vaccine (Oka-Strain) in Children with Chronic Liver Disease

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**SUMMARY** **Varicella infection may cause significant morbidity and mortality especially in immunocompromised persons. Children with chronic liver disease who undergo liver transplantation and need long term immunosuppressive therapy are at risk to acquire the infection. Twenty-nine children (aged 1-12 years) with chronic liver disease were enrolled to receive one dose of live attenuated varicella vaccine (Oka-strain). During the 16-week follow-up period, no vaccine-related serious adverse events were reported. Seroconversion rates at 8 weeks post vaccination were 100%. Geometric mean titer (GMT) values and seropositive rates at 16 weeks tended to relate to the clinical severity of liver disease. This study demonstrates that varicella vaccine is safe and immunogenic in children with chronic liver disease.**

Varicella is a common, highly contagious disease. The disease is usually mild in young children. However the severity of the disease increases with age. Furthermore varicella is one of the most important risk factors for severe, invasive, group A streptococcal disease.[1,2] The risk of complications and death attributable to varicella in adults is 10- to 20-fold higher than that of children. Despite the lower risks to severe morbidity and mortality among children, the burden of disease is the greatest since more than 90% of the cases occur in this age group.

Children with severe chronic liver disease or end stage liver disease are not likely to survive. Orthotopic liver transplantation (OLT) is currently an accepted procedure for the treatment of patients with end stage liver disease. After the transplantation, immunosuppressive drugs are required to prevent graft rejection. Varicella is one of the fatal infectious diseases found in immunocompromised persons but clinical disease does not develop in patients with serologic evidence or clinical history of varicella prior to transplantation.[3]

Special emphasis should be placed on immunization of susceptible older children and adults, because the likelihood of severe infection increases with age. Children with impaired humoral immunity including those with chronic liver disease may be immunized with varicella vaccine. However, according to the American Academy of Pediatrics, varicella vaccine should not be administered routinely to children who have cellular immunodeficiencies including leukemia, lymphoma, other malignancies affecting the bone marrow or lymphatic systems, congenital T-cell abnormalities and children receiving immunosuppressive therapy. Exceptions include children with acute lymphocytic leukemia, to whom vaccine may be given through a research protocol, and certain children infected with HIV.[4] Children infected with HIV may be at higher

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risk of morbidity from varicella and herpes zoster.

A live attenuated varicella vaccine (Oka-strain) was developed in Japan in the early 1970's[5] and is currently being used in many countries for the prevention of varicella infection in high-risk children. Varicella vaccine has been demonstrated to be very effective. Controlled, clinical trials demonstrated varicella vaccine to be 70% to 90% effective in preventing varicella infection and more than 95% effective in preventing severe varicella disease.[6,7] An efficacy study of 148 children performed during an outbreak of varicella in a childcare center in DeKalb County, GA, found that varicella vaccine was 86% effective in preventing varicella and 100% effective in preventing moderate to severe clinical disease. Varicella was less severe and resulted in fewer days of absence from the day care center among immunized patients in comparison with nonimmunized cases.[8] "Breakthrough" cases following exposure to wild-type varicella-zoster virus (VZV) occurs in about 1% to 4% of vaccinees per year, and the rate does not seem to increase with length of time after immunization.[9] However, the disease is usually of short duration, mild with fewer than 50 lesions and low-grade or no fever. The wild type virus can be differentiated from the vaccine virus type Oka strain by the restriction fragment length polymorphism.[10]

In a study of 68 pediatric renal or hepatic transplant candidates without natural immunity vaccinated with a live attenuated varicella vaccine (Oka-strain), seroconversion was evaluated at 30 and 90 days intervals post vaccination by ELISA and confirmed by immunofluorescence antibody to membrane antigen (FAMA).[11] Thirty days after vaccination, 50 percent of vaccinees were antibody positive by ELISA and 67% of nonresponders by ELISA were positive by FAMA (total seroconversion 30 days after vaccination was 64.7%). The second vaccination was done in nonresponders with negative ELISA and this increased the seroconversion rate to 73.5 % by the end of the study. There were no local or generalized adverse events reported or diagnosed. No intrafamilial varicella infection was reported.

Chronic liver disease such as biliary atresia usually progresses to cirrhosis or end stage liver disease and eventually liver transplantation may be needed. This study is therefore designed to evaluate the immunogenicity and reactogenicity of a single dose of live attenuated varicella vaccine (Oka-strain) in children with different stages of chronic liver disease .

## MATERIALS AND METHODS

### Study design

This was an open study. The protocol was approved by the Ethical Committee, Faculty of Medicine, Chulalongkorn University, Bangkok. All parents/guardians had been informed about the objective, risks and benefits of the study. Written consents were obtained prior to the study.

### Vaccine

The Oka strain varicella vaccine was developed by M. Takahashi (Osaka University, Japan), who isolated the virus from a healthy Japanese boy with typical varicella.[12] The vaccine master seed was obtained by propagating the virus in human embryonic lung cells, in guinea pig embryonic fibroblasts, and finally in WI 38 human diploid cells. The manufacturer's seed lot and working seed lot were obtained by further passages in MRC-5 human diploid cells.[13]

The commercially available Oka strain varicella vaccine (Varilrix™, SmithKline Beecham Biologicals, Belgium) was used in this study. The vaccine is provided as a lyophilized pellet, which is reconstituted before injection with sterile water provided by the manufacturer. One dose of the reformulated vaccine (0.5 ml) also contains a maximum of 25 μg of neomycin sulfate as a preservative and stabilizing ingredient. The vaccine used in this study was at a titer of ~ $10^4$ pfu/dose.

### Population study

A total of 29 children with chronic liver disease aged 1-12 years, who had no previous history of varicella/zoster infection or varicella immunization were enrolled in the study. Children with any known significant contact with anyone with varicella or herpes zoster during the preceding 4 weeks, receiving immunosuppressive therapy except topical steroid, with a history of neomycin sensitivity or allergic reactions to any previous vaccinations, or any acute febrile illness at the time of vaccination were excluded from the study. No immune globulin or blood product was allowed 3 months prior to varicella vaccination.

The children were divided into 3 groups according to the severity of illness. Groups I, II and III represented patients who still functioned normally, those who required con-

tinuous medical care as out patients and patients who needed multiple hospital admissions, respectively. The clinical diagnosis of these subjects is summarized in Table 1.

At the beginning of the study, all vaccinees who met the criteria were subjected to physical examinations, a collection of a pre-vaccination blood sample and they were injected subcutaneously one dose of vaccine in the upper left arm. Diary cards were provided for all subjects. The subjects' parents/guardians were instructed to record any local or systemic reactions such as fever, skin rash on the dairy card for 6 days after vaccine administration.

At 8 and 16 weeks post vaccination, any adverse events including rash occurring during the study period were reviewed with the investigator and the diary cards were collected. A post-vaccination blood sample for varicella antibodies was collected each time as well.

### Laboratory assays

Venous blood samples for serology testing were collected and processed so as to provide 0.5 ml of well-separated serum free of hemolysis contaminants. The serum samples were stored at -20°C and assayed for anti-VZV levels using the commercial immunofluorescence assay (IFA) produced by Hemagen Diagnostics, VIRGO Products Division, Columbia, Maryland (VIRGO VZV/IgG) with some modifications. The modifications were that the IFA test was performed with an in-house buffer (PBS, pH 7.4) and the first dilution was set at 1:4 instead of 1:8. The fluorescent antibody to membrane antigen (FAMA) test is generally considered the reference assay for varicella serology[14] but it is cumbersome despite improvements.[15]

The varicella titers were expressed as the reciprocal of the highest dilution that gave a positive result. Seroconversion was defined as the appearance of antibodies in the serum of subjects who were seronegative before vaccination (i.e., IFA titer < 4 before vaccination to ≥ 4 in the postvaccinal serum sample).[16] Results were considered invalid if the specimens contained artifacts which prevented accurate readings. Those invalid results were omitted from the analysis.

**Table 1** Clinical diagnosis of the subjects

| Diagnosis | Number |
|---|---|
| Biliary atresia | 18 |
| Infantile cholestasis | 4 |
| Congenital hepatic fibrosis | 2 |
| Extrahepatic portal hypertension | 1 |
| Biliary hypoplasia | 1 |
| Alagille syndrome | 1 |
| Wilson disease | 1 |
| Non cirrhotic portal fibrosis | 1 |
| **Total** | 29 |

**Table 2** Seropositive rate and geometric mean titers among children with chronic liver disease

| Group | n | Age (years) Mean ± SD | Seropositive rate | | GMT (95% CI) | |
|---|---|---|---|---|---|---|
| | | | Week 8 | Week 16 | Week 8 | Week 16 |
| 1 | 7 | 3.11 ± 3.05 | 100 (7/7) | 100 (6/6) | 105 (36-304) | 50 (23-111) |
| 2 | 10 | 4.74 ± 3.54 | 100 (8/8) | 88.9 (8/9) | 181 (84-386) | 34 (12-98) |
| 3 | 8 | 5.14 ± 4.03 | 100 (5/5) | 60 (3/5) | 84 (8-863) | 21 (0.9-459) |
| **Total** | 25 | 4.39 ± 3.51 | 100 (20/20) | 85 (17/20) | 119 (64-219) | 39 (20-77) |

Group 1 = patients who still functioned normally
Group 2 = patients who required continuous medical care as out patients
Group 3 = patients who needed multiple hospital admissions
*invalid results- 1 patient in group 2 at week 16; 3 patients in group 3 at weeks 8, 16
**lost visit- 1 patient in group 1 at week 16; 2 patients in group 2 at week 8

**Statistical methods**

The incidences of all recorded adverse events were calculated from all immunized subjects. Observed unsolicited symptoms and serious adverse events were reported descriptively. Geometric mean anti-varicella titers (GMTs) with 95% confidence intervals and seropositivity rate for each visit and each group were calculated. GMTs and the seropositive percentage were calculated for initially seronegative subjects. GMTs of specific varicella antibodies were calculated by log transformation of positive titers and by taking the antilog of the mean of these transformed values (titers < 4 were given an arbitrary value of 2).

## RESULTS

Twenty nine children were recruited into the study (14 males, 15 females). The mean age of the subjects was 4.38 years (range 1-12). Three children found to have varicella antibodies at the first visit were excluded only from the immunogenicity analysis. One girl died from massive upper gastrointestinal bleeding 10 days after receiving the vaccine. Thus the number of children included in the immunogenicity analysis was 25.

The children were divided into 3 groups according to the severity of illness. There were 7, 10 and 8 subjects in groups 1-3, respectively. Two patients in group 2 and 1 patient in group 1 were lost to follow up at week 8 and 16, respectively. The seropositivity rate and GMTs are presented in Table 2. Seropositivity rates at week 16 tended to relate to the severity of clinical illness.

Four children (14%) reported low-grade fever associated with upper respiratory tract infections ranging from 6 hours to 10 days after the vaccination. No local reactions (pain, redness or papulovesicular rash at the injection site), or serious adverse events related to the vaccine were reported.

## DISCUSSION

According to this study, we established that varicella vaccine is safe for children with chronic liver disease. The only adverse event reported was low-grade fever after vaccination. After thorough reviewing of the diary cards, this group of patients had symptoms associated with upper respiratory tract infections which were considered unrelated to vaccination. One child died from severe upper gastrointestinal bleeding after vaccination which was not related to the vaccine.

According to the American Academy of Pediatrics, the reactions from varicella vaccine are generally mild and occur with an overall frequency of approximately 5% to 35%.[4] Approximately 20% of immunized persons experienced minor injection site reactions (e.g. pain, redness, swelling). Approximately 3% to 5% of immunized children developed a localized rash, and an additional 3% to 5% developed a generalized varicella-like rash. These rashes typically consist of 2 to 5 lesions and may be maculopapular rather than vesicular; lesions usually appear 5 to 26 days after immunization. However, varicella-form rashes that occur within the first 2 weeks after varicella immunization in immunocompromised persons are due to wild-type VZV.[17] Although a temperature higher than 38.9°C (102°F) has been observed from 1 to 42 days after immunization in 15% of healthy immunized children, fever also occurs in a similar percentage of children receiving placebo and is not considered to be a significant adverse event of immunization.[7] A temperature higher than 37.8°C (100°F) has been reported in 10% of adolescents and adults who are immunized with the vaccine. Serious adverse events, such as encephalitis, ataxia, erythema multiforme, Stevens-Johnson syndrome, pneumonia, thrombocytopenia, seizures, neuropathy, and death, have been reported rarely in temporal association with varicella vaccine.[4] In some cases, wild-type VZV or another causal agent has been identified. In most cases, data are insufficient to determine a causal association.

Eight weeks post vaccination, all subjects in our study had seroconversion and had a good immune response (GMT 119; 95% CI 64-219). Regarding the seropositivity rate 16 weeks post vaccination of the three groups, the data showed that the first group remained 100 percent of seropositive rate whereas the seropositivity rate of the second group decreased to 88.9 percent and of the third group to 60 percent. The GMT at 16 weeks among the clinically less severe group was greater than the GMT of the clinically more severe group. Considering the GMT among those 3 groups of patients, we found that their GMT at 16 weeks was less than that at 8 weeks. These data suggested that the persistence of antibodies tended to relate to the clinical severity of chronic liver disease. In healthy children, the antibodies persisted for at least 6 years after receiving the same kind of varicella vaccine that we used in our study.[6]

The prevalence of malnutrition among children with end stage liver disease is high[18] and therefore immunocompetence in these patients was investigated. Hypoalbuminemia was common in all groups, with 66% (of those with chronic disease) having concentrations below 35 g/dl. Lymphopenia was equally common, 65% of patients with fulminant hepatic failure (FHF) had counts below 1,000 cells/mm$^3$. There were significant links between hypoalbuminemia, lymphopenia and immunoincompetence.[19]

In this study, unfortunately, there were not enough data to evaluate the invalid test results for varicella antibodies which were found mostly in the third group. The reason for this high incidence of invalid results in the third group was assumed to be that those patients had the most advanced clinical illness and therefore high plasma total lipids, low plasma albumin and high bilirubin. The effect of these factors might interfere with the testing technique.

Varicella can cause severe disease in immunocompromised children, like post liver transplant patients, who are on immunosuppressive drugs. Patients recently treated with high-dose prednisone and cyclosporine may suffer visceral dissemination resulting in deaths. Thus children in an early stage of chronic liver disease should be considered to receive varicella vaccine, before reaching end stage liver disease and prior to liver transplantation.

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## REFERENCES

1. Centers for Disease Control and Prevention. Outbreak of invasive group A *Streptococcus* associated with varicella in a childcare center: Boston, Massachusetts, 1997. MMWR Morb Mortal Wkly Rep 1997; 46: 944-8.
2. Laupland KB, Davies HD, Low DE, *et al.* Invasive group A streptococcal disease in children and association with varicella-zoster virus infection. Pediatrics 2000; 105: E60.
3. McGregor RS, Zitelli BJ, Urbach AH, *et al.* Varicella in pediatric orthotopic liver transplant recipients. Pediatrics 1989; 83: 256-61.
4. American Academy of Pediatrics. Committee on Infectious Diseases. Varicella vaccine update. Pediatrics 2000; 105; 136-41.
5. Takahashi M. Clinical overview of varicella vaccine: Development and early studies. Pediatrics 1986; 78 (suppl): 736-41.
6. Kuter BJ, Weibel RE, Guess HA, *et al.* Oka/Merck varicella vaccine in healthy children: Final report of a 2-year efficacy study and 7-year follow-up studies. Vaccine 1991; 9: 643-7.
7. Weibel RE, Neff BJ, Kuter BJ, *et al.* Live attenuated varicella virus vaccine: efficacy trial in healthy children. N Engl J Med 1984; 310: 1409-15.
8. Izurieta HS, Strebel PM, Blake PA. Postlicensure effectiveness of varicella vaccine during an outbreak in a childcare center. JAMA 1997; 278: 1495-9.
9. Clements DA. Modified varicella-like syndrome. Infect Dis Clin North Am 1996; 10: 617-29.
10. LaRussa P, Steinberg S, Arvin A, *et al.* Polymerase chain reaction and restriction fragment length polymorphism analysis of varicella-zoster virus isolates from the United States and other parts of the world. J Infect Dis 1998; 178 (Suppl 1): S64-6.
11. Giacchino R, Marcellini M, Timitilli A, *et al.* Varicella vaccine in children requiring renal or hepatic transplantation. Transplantation 1995; 60: 1055-6.
12. Takahashi M. Development and characterization of a live varicella vaccine (Oka strain) Biken J 1984; 27: 31-6.
13. D'Hondt E, Berge E, Colinet G, Duchene M, Peetermans J. Production and quality control of the Oka-strain live varicella vaccine. Postgrad Med J 1985; 61(suppl 4): 53-6.
14. Williams V, Gershon A, Brunell PA. Serologic response to varicella-zoster membrane antigens measured by indirect immunofluorescence. J Infect Dis 1974: 130: 669-72.
15. Zaia JA, Oxman MN. Antibody to varicella-zoster virus-induced membrane antigen: immunofluorescence assay using monodisperse glutaraldehyde-fixed target cells. J Infect Dis 1977; 136: 519-30.
16. Meurice F, De Bouver JL, Vandevoorde D, Woods S, Bogaerts H. Immunogenicity and safety of a live attenuated varicella vaccine (Oka/SB Bio) in Healthy Children. J Infect Dis 1996; 174: S324-9.
17. Feder HM Jr, La Russa P, Steinberg S, Gershon AA. Clinical varicella following varicella vaccination: don't be fooled. Pediatrics 1997; 99: 897-9.
18. de la Rubia MA, Serrano Corredor S, Lopez Soriano F, *et al.* The assessment of the nutritional status of patients in the terminal stage of liver disease who are candidates for orthotopic liver transplantation. Nutr Hosp 1994; 9: 163-9.
19. O'Keefe SJ, El-Zayadi AR, Carraher TE, Davis M, Williams R. Malnutrition and immuno-incompetence in patients with liver disease. Lancet 1980; 2: 615-7.