

BRIEF REPORTS

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## Evaluation of Immunogenicity and Tolerability of a Live Attenuated Hepatitis A Vaccine in Indian Children

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*An open non comparative study of a live attenuated H2 strain Hepatitis A vaccine of Chinese origin was carried out in 143 healthy Indian children aged 1 to 12 years (mean age 4.87 + 2.76 years; 88 boys, 55 girls). At baseline, all were negative for IgG HAV antibodies and had normal hematological and biochemical indices. Two months after a single dose of the vaccine (given subcutaneously), 137 children (i.e. 95.8%) developed protective antibodies of IgG >20 mIU/mL. The hematological and biochemical parameters remained within normal limits. There were no adverse events in any except mild fever in one child. In conclusion, live attenuated H2 strain Hepatitis A vaccine in a single dose was found to be immunogenic and safe in Indian children.*

**Key words:** *Immunogenicity, Live attenuated hepatitis A vaccine, Tolerability.*

**H**EPATITIS A Virus (HAV) has a worldwide distribution, and accounts for more than 1.4 million cases of viral hepatitis annually(1). In recent years, India and other transitional economies are showing a significant epidemiological shift of HAV infections from high endemicity to intermediate endemicity(2,3). Increasing numbers of older children and adults,

especially in urban 'developed' areas, are now susceptible to HAV with considerable increase in morbidity, costs and mortality related to the disease. Further, such areas (of transitional endemicity) are prone to recurrent and explosive epidemics of the disease as seen in Shanghai (China) and Kerala (India)(4,5). As such, WHO has now recommended hepatitis A vaccination programs in areas of intermediate

endemicity either as mass campaigns or as part of 'routine' immunization schedules(1). Large scale immunization programs in China, Israel, and in some states of USA have shown a remarkable reduction in the prevalence of the disease, implying that vaccination is a very effective public health tool in the control and potential elimination of the disease(6-8). The Indian Academy of Pediatrics (IAP) too has recently recommended hepatitis A vaccine as an 'additional' vaccine especially for children from higher socioeconomic groups in India(9).

Two kinds of hepatitis A vaccines are available (*i*) inactivated vaccines and (*ii*) live attenuated vaccine. Inactivated vaccines have been used worldwide since 1991, and are shown to be safe and effective with long lasting immunity(10). However, the prevailing costs of these vaccines precludes their widespread use in 'routine' schedules in India. As such, live attenuated vaccines derived from H2 strains are of great interest as they can be produced comparatively cheaply and in large quantities(6). These vaccines have been used successfully in millions of Chinese now for over a decade with promising results in primary prevention of HAV infections as well as in control of epidemics.

This study, which is the first experience of the vaccine outside China, was aimed at documenting the safety and efficacy of the H2 strain derived live attenuated hepatitis A vaccine in healthy Indian children beyond the age of one year.

### Subjects and Method

This study was carried out from August to December 2004. It was an open labelled, non-comparative study. After an informed consent, healthy subjects between the ages of 1-12 years who had not received hepatitis A vaccine were screened for anti-HAV antibodies (IgG), and routine hematology (Hb, complete blood

count) and biochemical liver profile (ALT, AST, Bilirubin). Children with negative anti-HAV antibodies and normal hematological and biochemical parameters were eligible for enrollment. Each enrolled child was given a single dose of 1ml of reconstituted freeze dried, live attenuated hepatitis A vaccine ('Zhepu', Zhejiang Pukang Biotechnological Company Ltd., China), subcutaneously over the deltoid muscle of the upper arm. Subjects were monitored closely for 1 hour following vaccination for immediate local or systemic events. Symptom diary cards were given to parents to note down any abnormal symptoms during the subsequent two months, and they were instructed to visit the hospital in case of any unexpected medical event. A repeat blood sample was taken two months after vaccination to estimate anti-HAV antibodies and hematology and biochemistry parameters. All blood samples were sent to and assessed by an independent laboratory (Ranbaxy Laboratories, Mumbai). Seroprotection was defined as an anti-HAV antibody level of IgG  $\geq$  20 mIU/mL after vaccination. Immunogenicity was estimated by determining the proportion of subjects who were seroprotected following vaccination. Vaccine safety and tolerability was assessed on the basis of (*i*) adverse events reported anytime after the administration of vaccine upto the completion of the study (2 months for each subject), and (*ii*) change in hematological or biochemical parameters.

This study was approved by the hospital ethical committee and was carried out as per ICH-GCP guidelines.

### Results

One hundred ninety-eight children aged 1-12 years were screened for inclusion criteria. 54 children (mean age  $7.8 \pm 2.5$  years) were positive for IgG HAV antibodies and were excluded. The remaining 144 children (mean

age  $4.9 \pm 2.8$  yrs) were negative for IgG HAV antibodies and were enrolled for the study. 143 (boys = 88, girls = 55) of these completed the trial and one was lost to follow up.

#### *Immunogenicity of the vaccine*

Whereas, all 143 children had non detectable levels of IgG HAV antibodies at baseline, 137 (*i.e.*, 95.8%) developed protective antibodies of IgG >20 mIU/mL by the end of two months. The distribution of children as per their antibody titers achieved is seen in Fig. 1.

#### *Safety and tolerability*

There were no immediate or delayed reactions and no serious adverse events following vaccination in any of the study children. The only adverse event probably related to the vaccine was mild fever, reported in one child. The fever lasted a few hours and subsided without any treatment.

Hematological and biochemical parameters at baseline and at the end of the trial remained within normal limits in all the children (Table I).

#### **Discussion**

The present study demonstrates the

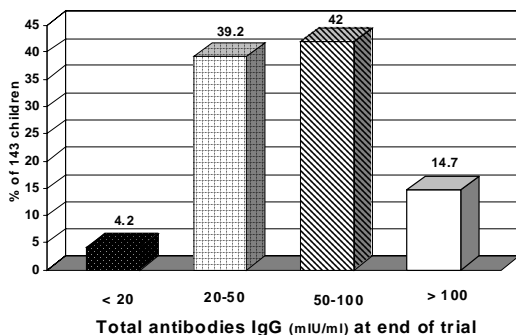


Fig. 1. Immunogenicity of live attenuated hepatitis A Vaccine 2 months after vaccination - IgG antibodies (mIU/mL).

remarkable safety and immunogenicity of the H2 strain derived live attenuated vaccine ('Zhepu', Zhejiang Pukang Biotechnological Company Ltd., China) in Indian children. The tolerance was excellent with no serious adverse drug reaction in any of the 144 children vaccinated. Of the 143 children evaluated, 137 (95.8%) developed protective antibodies (IgG >20 mIU/mL) against HAV by the end of 2 months, following a single subcutaneous injection of the vaccine. Long term follow up of these children has been planned to check on the persistence of antibodies. However, early results of the study are by and large similar to the Chinese studies by Mao, *et al.*(11) and Zuang, *et al.*(6).

Earliest attempts to develop a live attenuated vaccine were made in 1980. Success followed the isolation of a strain from the feces of a 12-year-old boy with HAV, near Hangzhou, China(12). This strain was further attenuated with several passages through cell cultures of newborn monkey kidney and human lung diploid fibroblasts. The attenuated vaccine was shown to be genetically stable, immunogenic in monkeys and nontransmittable by the oral route. The vaccine was licensed for human use in 1992, and since then has undergone extensive field trials in China in children and in adults(6,13). The vaccine induces not only neutralizing antibody but also cell mediated immune response suggesting the possibility of long term (life long?) immunity(14). Cohort studies by Zuang, *et al.* at 2 months, 10 years and 15 years after vaccination with a single dose showed persistence of protective antibody levels in 98.6%, 80.2% and 81.3% respectively(14). There were no major side effects and no elevation in serum aminotransferases were noted. In some areas of China (Shanghai), Hebei and Guanxi mass vaccination programs with live vaccines have led to over 95%

**TABLE I**—Hematological and Biochemical Parameters at Baseline and at End of Trial (2 months)

Parameters	Baseline		End of trial	
	Mean	(SD)	Mean	(SD)
Hemoglobin g%	12.3	1.5	11.7	1.5
WBC 10 <sup>3</sup> /cmm	10.6	3.2	10.3	3.3
Platelet count 10 <sup>3</sup> /cmm	371.4	94.6	338.1	89.9
AST U/L	31.9	10.4	32.6	8.6
ALT U/L	17.4	9.3	15.6	5.4
Bilirubin mg/dL	0.4	0.2	0.5	0.2

reduction in hepatitis A and no cases of HAV have been reported since 1999(6,13,14). These studies have also suggested that subclinical infections may actually act as boosters for the vaccine. The possibility of subclinical infections in our study children after receiving the vaccine, seems unlikely in the absence of elevation of the liver enzymes. H2 strain live vaccine has compared well in safety and immunogenicity studies with other HAV vaccines *viz.*, LA-1 derived live vaccine and an inactivated vaccine(13,15).

H2 strain derived live attenuated HAV vaccine has been recently introduced in India as Biovac A (Wockhardt). The obvious advantages of a single dose schedule, possible life long immunity, good tolerance and relatively lower costs make it an attractive option to the vaccines available in India for prevention and control of hepatitis A.

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*Contributors:* SB, AB, ZM and RJ were responsible for designing the study. JC was primarily responsible for day to day conduct of study. Shr. Bhu and AB

carried out statistical analysis. SB and ZM coordinated the study and drafted the paper. AP supervised all aspects of the study. SB will act as guarantor for the manuscript.

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*Competing Interests:* Dr. Zinobia Madan, Dr. Rasendrakumar Jha and Dr. Bhure are paid employees of Wockhardt India Ltd., Mumbai.

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### Key Message

- A live attenuated hepatitis A vaccine with extensive experience in China is now shown to be safe and effective in Indian children.

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