Granulocyte macrophage colony stimulating factor augmented hepatitis B vaccine protocol for rapid seroprotection in voluntary kidney donors

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Received September 9, 2003

Background & objectives: Conventional hepatitis B vaccine protocols do not provide rapid seroprotection against hepatitis B. This randomized controlled trial was carried out to investigate the efficacy of granulocyte macrophage-colony stimulating factor (GM-CSF) augmented double-dose vaccine protocol in voluntary kidney donors prior to donor nephrectomy.

Methods: A total of 54 kidney donors, who had no history of hepatitis B infection, hepatitis B vaccination and tested negative for anti-HBs and anti-HBc antibodies were randomly allocated to the control or test groups. GM-CSF (300 µg) was administered subcutaneously on day 0, followed by 40 µg of recombinant hepatitis B vaccine intramuscularly on the same deltoid on day 1. The control group received only 40 µg of intramuscular hepatitis B vaccine. Anti-HBs titres were measured at the end of 4 wk.

Results: Of the 54 donors studied, there was a significant (P<0.003) seroconversion in the GM-CSF group (82%) compared to the control group (37%), after a single immunization with double-dose recombinant hepatitis B vaccine by 4 wk. Minor side effects such as fever in four patients and myalgia in three were noticed.

Interpretation & conclusion: GM-CSF augmented double-dose hepatitis B vaccine could be used in unvaccinated patients when a rapid response is desired.

Key words GM-CSF - hepatitis B - vaccine augmentation

Voluntary kidney donors get about four weeks when essential tests prior to donor nephrectomy are conducted. Conventional hepatitis B vaccination does not result in protective antibody titres in kidney donors within such a short period. In endemic regions for hepatitis B virus particularly, in addition to universal precautions, it is important to protect these patients with an efficient vaccine protocol within the short interval prior to surgery. Recombinant human granulocyte macrophage colony stimulating factor (GM-CSF) has been shown as an adjuvant to improve the kinetics and magnitude of the immune response after vaccination1. GM-CSF is a haematopoetic growth factor, which affects the growth and function of granulocytes, macrophages, eosinophils and dendritic cells. It enhances neutrophil chemotaxis and phagocytic action of macrophages2,3.

Though the efficacy of GM-CSF in augmenting hepatitis B vaccine has been studied in normal subjects, patients with chronic renal failure and geriatric population, information is not available in voluntary kidney donors going for elective surgical procedure. This prospective randomized controlled study was undertaken to test the efficacy and safety of GM-CSF augmented double-dose
hepatitis B vaccination in inducing protective antibody titres as compared to non-augmented double-dose vaccine in voluntary kidney donors.

**Material & Methods**

The study was conducted at the Christian Medical College and Hospital, Vellore in southern India during October 2001 to June 2002. Consecutive prospective voluntary kidney donors who were found fit to donate kidneys were included after obtaining the informed consent. They were included in the control or test group based on a list of computer-generated random numbers. The study protocol was approved by the hospital ethics committee.

The inclusion criteria were as follows: (i) all consenting donors who were fit to donate a kidney after period of 4 wk; (ii) no history of hepatitis B infection or hepatitis B vaccination; and (iii) anti-HBs antibody and hepatitis B core antibody (anti-HBc) negative status.

Exclusion criteria were (i) donors likely to have less than a 4-wk vaccination-donor nephrectomy interval; (ii) current or past hepatitis B infection; (iii) history of side-effects with GM-CSF; and (iv) anti HBs and/or anti-HBc positivity.

The test group received 300 µg of GM-CSF (Leucomax, Novartis, Basle, Switzerland) subcutaneously on day 0 followed by 40 µg of recombinant hepatitis B vaccine intramuscularly on the ipsilateral deltoid on day 1. The control group received 40 µg of intramuscular hepatitis B vaccine only. Anti-HBs titre was measured at the end of 4 wk using AUSAB kit (Axsym, Abbot Labs, IL, USA). HBsAg was tested using a third generation ELISA kit (Hepanostika Uniform II - Organon Technika Netherlands, or MEIA-Axsym, Abbott Lab, USA) and anti-HBc was tested with CORE kit (Axsym, Abbott Labs, IL, USA). Donors were advised to continue the vaccination at one and six months after the first dose to ensure persistence of protective antibody levels.

Chi square test and Student t-test were used to determine difference in the two groups. Mann Whitney U test was done to study difference in titres between the two groups. P<0.05 was considered significant.

**Results & Discussion**

The study population consisted of 54 donors; 25 were males. The mean age was 37.9±9.3 yr. There was no significant difference in age between subjects in the test and the control groups, mean age in GM-CSF group being 37.3±10.7 and in control group 37.55±10.08 yr (Table). There were 27 donors in each group.

Thirty four voluntary kidney donors seroconverted with an anti-HBs titre of more than 10mIU/ml, 23 in the GM-CSF group seroconverted compared to the 11 from the control group, the difference was statistically significant (P<0.001). The median anti-HBs titres were 32 mIU/ml and 8 mIU/ml in the study group and controls respectively. The mean titre achieved in the GM-CSF group (42.12±36mIU/ml) was significantly (P<0.003) higher compared to 20.33±38.5 mIU/ml in the control group (Table). Minor side effects viz., fever in four patients and myalgia in three patients were noticed in the GM-CSF group.

Voluntary kidney donors need protective antibody levels against hepatitis B prior to donor nephrectomy to avoid possible infection with the virus. Conventional vaccine protocols do not provide protective antibody levels in 4 wk⁴. GM-CSF produces improvement in the kinetics and antibody response in human studies⁵,⁶. The ability of GM-CSF to stimulate hepatitis B vaccine in haemodialysis patients⁷-⁹ with a defect in antigen presenting cells and in nonresponders have been shown in earlier studies.

In the present study the efficacy of GM-CSF augmented double-dose vaccine protocol was evaluated. A 300 µg dose of GM-CSF was given subcutaneously 24 h prior to administering hepatitis B vaccine (40 µg)

**Table.** Comparison of parameters between test and control groups

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<tr>
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<th>Test group (n=27)</th>
<th>Control group (n=27)</th>
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<tr>
<td>Mean age (yr)</td>
<td>37.13±10.7</td>
<td>37.55±10.08</td>
</tr>
<tr>
<td>Sex (male:female)</td>
<td>11:16</td>
<td>14:13</td>
</tr>
<tr>
<td>Seroconversion n (%)</td>
<td>23 (82)*</td>
<td>11 (37)</td>
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<tr>
<td>Mean titres (mIU/ml)</td>
<td>42.12±36†</td>
<td>20.33±38.5</td>
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*P<0.001 compared to control group
†P<0.003 compared to control group
intramuscularly at the same site as GM-CSF, since GM-CSF is known to produce local inflammation at the site of injection and enhances dendritic cell maturation and migration. Confounding factors like previous hepatitis B infection, vaccination, and the remote possibility of subclinical hepatitis B infection were removed by excluding subjects having antibody to HBV core antigen.

The findings showed a significant seroconversion (82%) in the GM-CSF group as compared to the control group (37%) after a single immunization with double-dose recombinant hepatitis B vaccine. Some of the earlier studies however, have failed to demonstrate the adjuvant effect of GM-CSF or has shown only a marginal adjuvant effect. The effect of GM-CSF shown in this study may be due to the use of higher doses of GM-CSF and hepatitis B vaccine and the same site for vaccination.

The GM-CSF group had significantly higher titres of anti-HBs compared to the control group and it was well tolerated except for mild fever and myalgia, making the protocol applicable to similar situations elsewhere.

In conclusion, GM-CSF was found to be a safe and effective hepatitis B vaccine adjuvant. The potential of GM-CSF to augment double-dose hepatitis B vaccine could be used in unvaccinated patients when a rapid response is desired prior to elective surgical procedures and in situations where protective antibody levels are required for health care workers.

Acknowledgment

The authors acknowledge the financial support from Novartis, India.

References


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### SOME FORTHCOMING SCIENTIFIC EVENTS

<table>
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<th>Symposium/Seminar/Workshop Course/Conference (Date &amp; Place)</th>
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<tr>
<td>World Congress on Fertility &amp; Sterility (May 23-28, 2004; Montreal, Quebec, Canada)</td>
<td>June Viau/Scientific Program Coordinator, IFFS 2004 Congress Secretariat, 205 Viger Avenue West, Suite 201, Montreal QC H2Z 1G2, Canada Tel: 514-874-1998; Fax: 514-874-1580 e-mail: <a href="mailto:june@fa-events.com">june@fa-events.com</a> Web site: <a href="http://www.IFFS2004.com">www.IFFS2004.com</a></td>
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<td>National Workshop cum Symposium on Recent Developments in Haematology: An Indian Perspective (June 4-6, 2004; Kolkata)</td>
<td>Dr Prantar Chakrabarti, Organizing Secretary, Assistant Professor, Institute of Haematology &amp; Transfusion Medicine, 3rd Floor, M.C.H. Building Medical College, Kolkata, 88 College Street, Kolkata 700073, India e-mail: <a href="mailto:prantar@ihtmindia.org">prantar@ihtmindia.org</a> <a href="mailto:prantan@vsnl.net">prantan@vsnl.net</a></td>
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<td>XII International Congress on Endocrinology (August 31-September 4, 2004; Lisbon, Portugal)</td>
<td>KIT GmbH, Convention and Incentive Organization Kurfürstendamm 71, D-10709 Berlin, Germany Tel: ++49-30-246-03-301; Fax: ++49-30-246-03-310 e-mail: <a href="mailto:info@ice2004.com">info@ice2004.com</a> Web site: <a href="http://www.kit.de">http://www.kit.de</a></td>
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<td>Golden Jubilee Conference of The Journal of Postgraduate Medicine (JPGM GOLDEN) (September 23-26, 2004; Mumbai)</td>
<td>Dr Nithya Gogtay, Organizing Secretary, Department of Clinical Pharmacology Seth GS Medical College &amp; KEM Hospital, Parel, Mumbai 400012, India Tel: 91-022-24133767/24174420 Fax: 91-022-24143435 e-mail: <a href="mailto:nithvagogtav@gsmc.edu">nithvagogtav@gsmc.edu</a></td>
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