Original Article

Sci-B-VacTM Vs ENGERIX-B Vaccines for Hepatitis B Virus in Patients with Inflammatory Bowel Diseases: A Randomised Controlled Trial

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Abstract

Background and Aims: Response rate to second-generation hepatitis B virus vaccines is relatively low in patients with inflammatory bowel diseases compared with the general healthy population. We compared the efficacy and safety of a third- vs a second-generation hepatitis B virus vaccine in a group of patients with inflammatory bowel diseases treated with immunosuppressive medications.

Methods: Prospective, randomised, single-blind, controlled study. Eligible patients were randomly assigned to receive one of two vaccines, ENGERIX-B or Sci-B-Vac. The vaccines were administered in three doses at 0, 1, and 6 months. The primary endpoint was defined as the titre of anti-hepatitis B S [HBs] antibodies following the standard three-dose hepatitis B virus vaccination schedule.

Results: A total of 72 patients complied with study protocol [37 and 35 patients in the ENGERIX-B and Sci-B-Vac groups, respectively]. Overall, 75% of the cohort seroconverted. The primary endpoint was met in 81.1% in the ENGERIX-B group and 68.6% in the Sci-B-Vac group [$p = 0.22$]. Patients in the Sci-B-Vac group showed a statistically significant decreased seroconversion rate compared with the ENGERIX-B group, with use of tumour necrosis factor [TNF] alpha inhibitors [$p = 0.03$], and higher degree of disease activity [$p = 0.03$].

Conclusions: Overall seroconversion rate in our cohort was higher than in previous reports in the literature, possibly due to a low disease activity state in the majority of participants. Third-generation hepatitis B virus vaccines showed no apparent advantage over standard of care vaccine in this patient group.

Key Words: Hepatitis B virus vaccine; inflammatory bowel disease; immunosuppression

1. Introduction

In the past decade, the treatment of inflammatory bowel disease [IBD] has changed dramatically with the growing use of immunomodulators and the advent of novel biological agents.

Biological agents administered as stand-alone treatment or in combination with other immunosuppressants have become the backbone of therapy.¹ However, the growing use of immunomodulators came with the inherent risks of immunosuppression. Infections complicating

the course of IBD have become a major health concern in this population, and especially in those maintained on long-term therapy with one or more immunosuppressants.^{2,3}

Hepatitis B virus [HBV] affects more than 350 million people worldwide and is one of the preventable infections which can complicate the course of IBD.⁴ Indeed, a number of case reports and small case series published in the past decade have reported symptomatic and, at times, severe HBV flares in inactive HBV carriers with IBD receiving tumour necrosis factor [TNF]-alpha inhibitors.^{5,6,7} Several professional societies have issued recommendations regarding screening and management of HBV infection in immunosuppressed patients.^{8,9,10} In seronegative patients, the European Crohn's and Colitis Organisation's ECCO consensus statement included a universal recommendation for HBV vaccination.

Second-generation HBV vaccines such as ENGERIX-B contain the recombinant major S antigen component of the virus. Although these vaccines are immunogenic in over 95% of the general adult population,¹¹ response rates in subgroups of immune-compromised patients are lower. In these patients, a three double-dose ENGERIX-B injection protocol has been shown to improve seroconversion rates.^{12,13} This strategy recently applied in IBD patients showed improved response rates to HBV vaccine compared with a standard dose vaccination schedule. However, only 75% of IBD patients in the double-dose arm achieved protective hepatitis B S [HBs] antibody level following vaccination with this protocol.¹⁴

Sci-B-VacTM is a third-generation HBV vaccine that contains epitopes of the major S antigen as well as the minor pre-S1 and pre-S2 envelope proteins. This tri-epitope vaccine administered in a three standard-dose vaccine schedule was shown to induce higher response rates than ENGERIX-B in the general population¹⁵ as well as in 'difficult to vaccinate' subgroups such as healthy non-responders¹⁶ and patients with end-stage kidney disease.¹⁷

The primary objective of our study was to compare the efficacies of Sci-B-VacTM and ENGERIX-B in a group of IBD patients treated with one or more immunosuppressant medications.

2. Methods

2.1. Study population

Patients from the IBD clinic of Soroka University Medical Center [SUMC], a tertiary care 1000-bed medical centre located in the southern part of Israel, were enrolled in the study. Inclusion criteria were: [i] male or female ≥ 18 years of age; [ii] evidence of Crohn's disease [CD] or ulcerative colitis [UC] as diagnosed by clinical, laboratory, imaging, and endoscopic criteria; [iii] seronegativity to hepatitis B S antigen [HBsAg], antibodies to hepatitis B core protein [anti-HBc], and antibodies directed against hepatitis B surface antigen [anti-HBs]; [iv] treatment with at least one immunosuppressive medication at the time of study initiation; and [v] written informed consent. We excluded [i] pregnant women; [ii] patients with previous HBV vaccination regardless of anti-HBs titre; [iii] history of a major acute medical event occurring within 30 days of enrolment in the study [eg acute myocardial infarction, stroke, pulmonary emboli, sepsis, or major trauma]; [iv] haematological or oncological malignancies occurring within 2 years before enrolment in the study; and [v] history of chronic diseases other than IBD associated with immune aberrations [eg connective tissue disease, cirrhosis, end-stage kidney disease, and HIV infection].

Immunosuppressive medications included systemic steroids [prednisone], purine analogues [purinethol and azathioprine], methotrexate, and TNF-alpha inhibitors [infliximab and adalimumab].

2.2. Study design

This was a single-blind randomised controlled trial. Both the patients and the laboratories performing the assays for this study were blinded to treatment assignment; however, physicians administering the vaccines were not. Eligible patients were randomised 1:1 to receive one of two vaccination regimens: either three intramuscular injections of recombinant ENGERIX-B vaccine [1.0 ml, 20 µg per dose; GlaxoSmithKline, UK], administered at 0, 1, and 6 months; or three intramuscular doses of the third-generation Sci-B-Vac vaccine, [1.0 ml, 10 µg per dose; SciGen, Israel] administered at 0, 1, and 6 months. Both vaccines were injected to the deltoid muscle.

Sci-B-Vac, a third-generation HBV vaccine, was selected for comparison with ENGERIX-B on the basis of results of previous studies showing its superior efficacy in subgroups of patients who failed to respond to a second-generation vaccine. Third-generation HBV vaccines are produced in mammalian cells as opposed to recombinant second-generation vaccines, which are yeast-derived. These vaccines contain glycosylated and non-glycosylated virus-like particles of the major S [similar to second-generation vaccines], as well as Pre-S1 and Pre-S2 regions.

2.3. Data collection

At study enrolment, demographic data and detailed information regarding baseline disease characteristics, disease activity scores (Crohn's Disease Activity Index [CDAI] and partial Mayo Clinic score [pMayo-Clinic] for CD and UC, respectively), past and current medications, and history of surgical interventions were collected from all participants. During each of the three-vaccine injection visits, CDAI/pMayo-Clinic activity score questionnaires were filled in, and data on any changes in type and dosage of IBD medications were collected.

Blood was drawn at baseline and at each vaccine administration visit for complete blood count [CBC], chemistry [glucose, electrolytes, liver function tests, albumin and total protein, creatinine and urea], and C-reactive protein [CRP] levels. Anti-HBs titres were measured 30 days following first and second doses of vaccine, and 1–3 months following the third vaccine dose.

2.4. Safety assessment

Patients were observed for at least 20 min following vaccine injection for the development of any life-threatening allergic reactions. In addition, participants were instructed to contact one of the study investigators to report the occurrence of adverse reactions in the days following vaccine injection. Patients were questioned regarding any possible adverse events during the study follow-up visits.

2.5. Primary endpoint

The primary endpoint was a titre of anti-HBs antibodies following standard three-dose schedule HBV vaccination.

2.6. Secondary endpoints

Secondary endpoints included the rate of development of protective anti-HBs antibodies following a third dose of either ENGERIX-B or Sci-B-Vac vaccines, defined as an antibody titre > 10 IU/ml 30 days following the third vaccination [adequate immune response]. In addition, we have assessed the rates of development of anti-HBs antibody titre > 100 mIU/ml 30 days following the first, second, and third vaccination doses [effective immune response], rates of development of anti-HBs antibody titre > 10 mIU/ml 30 days following the first and second vaccination doses, median anti-HBs antibody titre following

each vaccination dose, the rate of severe adverse events, and compliance rates with full vaccination protocol.

2.7. Sample size calculations, randomization, and statistical analysis

We hypothesised that the two treatments will result in different antibody titres. We estimated the log-transformed mean for antibody titre to be equal to 4, with coefficient of variation equal to 0.6. A sample size of 50 subjects in each group achieved a power of 80% to detect a ratio above 1.4 between the two vaccinated groups.

Data collected in this study were summarised using frequency tables, summary statistics, confidence intervals, and *p*-values as appropriate. We used chi-square tests for categorical variables, *t* test for normally distributed continuous variables, and Wilcoxon rank sum test for variables diverging from the normal distribution.

Randomisation was performed using a computer-generated randomisation list by the research pharmacy.

Analysis of primary and secondary endpoints was stratified according to number and type of immunosuppressive medications, as well as by presence or absence of disease remission at baseline.

Active disease was defined clinically as CDAI ≥ 150 or pMayo-Clinic ≥ 2 , for CD and UC respectively, and biochemically as CRP levels > 0.5 mg/dl for both diseases.

To assess the adjusted effect of vaccine type, linear regression models with log-transformed titres of anti-HBs antibody, inclusive of vaccine type, remission status at enrolment, and use of Tnf- α inhibitors, were applied.

Results were considered significant when the two-sided *p*-values were < 0.05 . Analyses were performed using the Statistical Program for Social Science [SPSS 21.0 for Windows; SPSS Inc., Chicago, IL, USA].

2.8. Ethical statement

Study protocol was approved by the local institutional review board and was conducted according to the Declaration of Helsinki and Good Clinical Practice Guidelines. The lead author and co-authors had access to the study data and all reviewed and approved the final manuscript.

3. Results

3.1. Population

A total of 117 patients from the Soroka Medical Center IBD clinic were assessed for eligibility between May 2012 and March 2013; 26 refused to participate, and 91 patients were enrolled and sent for screening after providing written informed consent. After exclusion of 17 patients, 73 participants were randomly assigned to receive either ENGERIX-B [$n = 37$] or Sci-B-Vac [$n = 36$] vaccines. Following the first vaccination dose, one patient from the Sci-B-Vac group reported having a positive urine pregnancy test, and was subsequently excluded from the trial. The remaining 72 patients complied fully with study protocol and were thus included in the final analysis [Figure 1].

Table 1 depicts baseline characteristics of patients in the ENGERIX-B and Sci-B-Vac arms. Mean age, gender, and ethnic distribution as well as background comorbidities and IBD duration were similar between the two groups. In both treatment arms, CD was the predominant IBD form [73% and 85.7% in the ENGERIX-B and Sci-B-Vac groups, respectively]. Calculated disease activity scores at the time of first vaccine injection were consistent with disease remission in the majority of patients in both vaccine arms [78.4% in ENGERIX-B group and 88.6% in Sci-B-Vac group; $p = 0.24$]. The proportion of patients with CRP values within the

Figure 1. Study flow chart.

Table 1. Baseline patient characteristics.

	ENERGIX-B <i>n</i> = 37	Sci-B-Vac <i>n</i> = 35	<i>p</i> -Value
Age [$\pm$ SD]	38.0 [$\pm$ 12.7]	37.6 [$\pm$ 14.5]	0.90
Male gender [%]	15 [40.5%]	21 [60%]	0.09
Ethnicity [%]			
Jews	33 [89.2%]	30 [85.7%]	0.65
Bedouins	4 [10.8%]	5 [14.3%]	
Comorbidities [%]			
Diabetes mellitus	2 [5.4%]	1 [2.9%]	0.58
HTN	2 [5.4%]	3 [8.6%]	0.59
Dyslipidaemia	5 [13.5%]	3 [8.6%]	0.50
Asthma	0	2 [5.7%]	0.14
COPD	0	1 [2.9%]	0.29
IBD type [%]			
Crohn's disease	27 [73%]	30 [85.7%]	0.18
Ulcerative colitis	10 [27%]	5 [14.3%]	
Number of IBD years	10.70 [$\pm$ 8.74]	9.59 [$\pm$ 7.17]	0.55
CRP median [IQR]	0.495 [0.16–1.23]	0.59 [0.13–1.26]	0.6
CRP mean [$\pm$ SD]	0.72 [$\pm$ 0.67]	1.39 [$\pm$ 2]	0.07
CDAI median [IQR]	52 [5–78]	35.1 [14–85.75]	0.85
pMayo-Clinic, median [IQR]	2 [0–5.25]	0 [0–1]	0.86
Remission according to CDAI and pMayo-clinic scores [first vaccination], [%]	29 [78.4%]	31 [88.6%]	0.24
Remission according to CRP levels [first vaccination] [%]	19 [51.4%]	15 [44.1%]	0.54
Type of immunosuppressive drugs taken [%]			0.44
Purine analogues [%]	22 [61.1%]	23 [69.7%]	
TNF-alpha inhibitors [%]	4 [11.1%]	2 [6.1%]	
Purine analogues + TNF-alpha inhibitors [%]	6 [16.7%]	3 [9.1%]	
Purine analogues + steroids [%]	2 [5.6%]	0 [0.0%]	
TNF-alpha inhibitors + steroids [%]	1 [2.8%]	1 [3%]	
Purine analogues + TNF-alpha inhibitors + steroids [%]	1 [2.8%]	3 [9.1%]	
Antimetabolites [methotrexate] [%]	0	1 [3%]	

SD, standard deviation; COPD, chronic obstructive pulmonary disease; IBD, inflammatory bowel disease; CRP, C-reactive protein; CDAI, Crohn's Disease Activity Index; TNF, tumour necrosis factor; IQR, interquartile range; HTN, hypertension.

normal range [< 0.5 mg/l] was considerably lower and comparable between the two groups [51.4% and 44.1% in the ENGERIX-B and Sci-B-Vac arms, respectively; $p = 0.542$]. Most participants in the ENGERIX-B and Sci-B-Vac vaccination groups were treated with only one immunosuppressive medication at any time point during the study period [70% and 72%, respectively]. TNF-alpha inhibitor regimens as either stand-alone treatment or in combination with other immunosuppressants were used in 32% and 26% of patients in the ENGERIX-B and Sci-B-Vac groups, respectively [$p = 0.44$].

3.2. Vaccination outcomes

Median anti-HBs titre following the third vaccine dose did not differ between the vaccination groups: 183.89 (interquartile range [IQR] 14.11–1000) in the ENGERIX-B group and 71.22 [IQR 8.15–619.52] in the Sci-B-Vac group. The ratio of mean log-transformed titres between ENGERIX-B and Sci-B-Vac groups was 1.18 (95% confidence interval [CI] 0.60–2.33, $p = 0.62$) [Figure 2a]. Overall, 75% of patients in our cohort achieved adequate immune response [AIR] following a three-dose vaccine regimen, whereas only 45% reached effective immune response [EIR].

At the lower cutoff level [AIR], seroconversion rates were not statistically significantly different in patients from the ENGERIX-B group compared with those from the Sci-B-Vac group [81.1% vs 68.6%, $p = 0.22$] [Figure 2b]. A similar and even stronger trend in seroconversion rates between the two arms was also observed for the [EIR] [56.8% and 37.1% in the ENGERIX-B and Sci-B-Vac groups, respectively, $p = 0.09$] [Figure 2c]. Notably, differences between the two arms in median anti-HBs titres as well as in seroconversion rates

became prominent only after completion of the three-dose vaccine schedule [Figures 2 a-c].

3.3. Subgroup analyses

The majority of patients in this cohort received purine analogue monotherapy [$n = 47$]. In this group, AIR rate was relatively high and comparable between the two groups [87 vs 83.3% in Sci-B-Vac and ENGERIX-B arms, respectively; $p = 1$]. However, response rates in patients treated with TNF-alpha inhibitors as either stand-alone treatment or when combined with other immunosuppressants [$n = 21$] were significantly lower in the Sci-B-Vac vs ENGERIX-B arms [22.2 vs 75%, respectively; $p = 0.03$]. At the EIR threshold, the difference between groups trended towards significance [22.2 vs 66.7% in Sci-B-Vac vs ENGERIX-B arms, respectively; $p = 0.08$] [Table 2].

Four patients in each arm were treated with systemic steroids at any time point during the study period [11.4 and 10.8% in the Sci-B-Vac and ENGERIX-B, respectively]; five at the time of the first vaccine injection and the rest after this time point. Daily dosage of prednisone varied between 15 and 45 mg, and the length of treatment was between 4 weeks and 7 months. Six out of the eight patients treated with steroids were also on TNF-alpha inhibitor therapy, whereas the other two were concomitantly treated with purine analogues. The difference in response rates to vaccination among patients treated with systemic steroids in the two arms was not statistically significant [Table 2].

In the subgroup of patients with clinically active disease [CDAI > 150 or pMayo-Clinic > 2 , $n = 12$], AIR was achieved in

Figure 2. Efficacy of Sci-B-Vac vs ENGERIX-B. a: Median anti-HBs titre levels. b: Rate of development of anti-HBs > 10 mIU/ml. c: Rate of development of anti-HBs > 100 mIU/ml. HBs, hepatitis B S.

a significantly lower proportion of patients in the Sci-B-Vac arm compared with the ENGERIX-B arm [25% vs 87.5%, respectively; $p = 0.03$]. Similarly, none of the patients with clinically active disease

in the Sci-B-Vac arm accomplished EIR at end of study, whereas 5/8 patients [62.5%] in the ENGERIX-B arm did [$p = 0.03$]. The differences in efficacy between the two vaccines were even more evident

when a CRP > 0.5mg/dl was used as a marker for disease activity. Approximately 89% of patients in the ENGERIX-B arm compared with 52.6% of patients in the Sci-B-Vac achieved AIR [$p = 0.01$], and only 1/19 [5.3%] vs 12/18 [66.7%] of patients accomplished EIR at end of study, in the Sci-B-Vac and ENGERIX-B arms respectively [$p < 0.001$] [Tables 3]. Overall, the only variable found to be a significant predictor of successful vaccination was the use of TNF-alpha inhibitors [20.4 vs 55.6% in responders vs non-responders, respectively; $p = 0.004$] [Table 4].

In linear regression models that included vaccine type, remission status at enrolment, and use of TNF-alpha inhibitors, no statistically significant difference was found between vaccines in achieving higher titre of antibody [$p = 0.42$]

3.4. Safety

In both study arms, no patient experienced any severe adverse events related to vaccine administration. Three patients were hospitalised due to Crohn's disease exacerbation during the study period, two from ENGERIX-B and one from Sci-B-Vac group. These admissions were not temporally associated with vaccine administration [occurring 2–3 months following the second dose of vaccine] and were thus not attributed to the vaccines.

4. Discussion

In this randomised controlled trial, we did not find a statistically significant difference in efficacy between Sci-B-Vac [a third-generation HBV vaccine] and the widely used ENGERIX-B vaccine, in a cohort of IBD patients being treated with immunosuppressive therapy. In fact, a possible trend was noticed towards superior efficacy of ENGERIX-B at the higher threshold of anti-HBs antibody level. Subgroup analysis showed that the difference in response rates between the two groups was mainly accounted for by the poorer

efficacy of Sci-B-Vac among patients treated with TNF-alpha inhibitors, either as stand-alone treatment or when combined with other immunosuppressants.

Individuals with IBD are considered to be at high risk for HBV reactivation, mainly due to the now common use of immunosuppressive and biological therapy for long periods. Routine screening and vaccination of patients with IBD are therefore recommended by the major gastrointestinal and liver professional societies. Unfortunately, accumulated data show a considerably lower efficacy of standard HBV vaccine in the IBD population compared with healthy subjects. Only two studies so far have explored the utility of modified HBV vaccination regimens in IBD patients. The first, by Gispert *et al.*, showed that an accelerated [0, 1, and 2 months] double-dose vaccine protocol led to a higher seroconversion rate compared with a standard-dose vaccine regimen.¹⁴ The second, by Cossio-Gil *et al.*, demonstrated a 50% response rate upon revaccination with a standard-dose vaccine, in IBD patients who initially failed to respond to the same vaccine regimen.¹⁸ In both studies, however, response rate in the superior treatment arm did not exceed 75%. In our cohort, the overall response rate was 75% and exceeded 85% in the ENGERIX-B arm. To the best of our knowledge, this is the highest response rate reported so far in the literature, in IBD patients vaccinated against hepatitis B. It should be stressed that these high response rates were achieved despite the fact that all the patients in this cohort were being treated with immunosuppressants. Improved vaccine efficacy in this study can be attributed to two main factors. First, younger age has been associated with improved response to HBV vaccine in the general population as well as in IBD patients.¹⁹ The mean age of patients in our study was 37, which is somewhat younger compared with that reported in similar studies. More importantly, the majority of patients in this cohort were in remission throughout the study period, as objectively assessed by disease activity scores. It has been suggested that active disease might impair

Table 2. Seroconversion rate according to therapeutic regimen.

Immunosuppressive regimen [%]	Seroconversion rate Anti HBs > 10 IU/ml			Seroconversion rate Anti HBs > 100 IU/ml		
	ENERGIX-B	Sci-B-Vac	<i>p</i> -Value	ENERGIX-B	Sci-B-Vac	<i>p</i> -Value
Purine analogues, <i>n</i> = 47	20/24 [83.3%]	20/23 [87%]	1	13/24 [54.2%]	11/23 [47.8%]	0.773
TNF-alpha inhibitors, with or without other medications, <i>n</i> = 21	9/12 [75%]	2/9 [22.2%]	0.03	8/12 [66.7%]	2/9 [22.2%]	0.08
Systemic steroids, <i>n</i> = 8	4/4 [100%]	1/4 [25%]	0.14	3/4 [75%]	1/4 [25%]	0.49
TNF-alpha inhibitors and/or systemic steroids, <i>n</i> = 23	11/14 [78.6%]	2/9 [22.2%]	0.013	9/14 [64.3%]	2/9 [22.2%]	0.089

HBs, hepatitis B S; TNF, tumour necrosis factor.

Table 3. Seroconversion rate according to disease activity state.

Immunosuppressive regimen [%]	Seroconversion rate Anti HBs > 10 IU/ml			Seroconversion rate Anti HBs > 100 IU/ml		
	ENERGIX-B	Sci-B-Vac	<i>p</i> -Value	ENERGIX-B	Sci-B-Vac	<i>p</i> -Value
Active disease according to CDAI ^a and partial Mayo Clinic ^b scores [%], <i>n</i> = 12	7/8 [87.5%]	1/4 [25%]	0.03	5/8 [62.5%]	0/4 [0%]	0.03
Active disease according to CRP levels mg/dl [%], <i>n</i> = 37 ^c	16/18 [88.9%]	10/19 [52.6%]	0.01	12/18 [66.7%]	1/19 [5.3%]	< 0.001

HBs, hepatitis B S; CDAI, Crohn's Disease Activity Index; CRP, C-reactive protein

^aActive disease according to CDAI score is defined as CDAI score ≥ 150 .

^bActive disease according to partial Mayo Clinic score is defined as Mayo Clinic score ≥ 2 .

^cActive disease according to CRP levels is defined as CRP ≥ 0.5 mg.

Table 4. Factors associated with non-response to HBV vaccination.

		Responders	Non-responders	<i>p</i> -Value
		<i>n</i> = 54	<i>n</i> = 18	
Age, [$\pm$ SD]		36.41 [$\pm$ 12.88]	41.83 [$\pm$ 14.86]	0.14
Male gender, [%]		27 [50%]	9 [50%]	1.00
Ethnicity, [%]	Jews	48 [88.9%]	15 [83.3%]	0.53
	Bedouins	6 [11.1%]	3 [16.7%]	
Comorbidities, [%]	Diabetes mellitus	3 [5.6%]	0	0.32
	Hypertension	3 [5.6%]	2 [11.1%]	0.42
	Dyslipidaemia	5 [9.3%]	3 [16.7%]	0.38
	Asthma	1 [1.9%]	1 [5.6%]	0.40
	COPD	1 [1.9%]	0	0.57
IBD type, [%]	Crohn's	41 [75.9%]	16 [88.9%]	0.24
	Ulcerative colitis	13 [24.1%]	2 [11.1%]	
Number of IBD years		10.34 [$\pm$ 7.80]	9.60 [$\pm$ 8.71]	0.73
Remission according to CDAI and partial Mayo Clinic [first vaccination] [%]		46 [85.2%]	14 [77.8%]	0.46
Remission according to CRP levels [first vaccination] [%]		27 [50.9%]	7 [38.9%]	0.37
Type of vaccination, [%]	ENERGIX-B	30 [55.6%]	7 [38.9%]	0.22
	Sci-B-Vac	24 [44.4%]	11 [61.1%]	
Received TNF-alpha inhibitors, [%]		11 [20.4%]	10 [55.6%]	0.004
Total number of drugs taken [%]	1	43 [79.6%]	12 [66.7%]	0.39
	2	9 [16.7%]	4 [22.2%]	
	3	2 [3.7%]	2 [11.1%]	

HBV, hepatitis B virus; SD, standard deviation; IBD, inflammatory bowel disease; CDAI, Crohn's Disease Activity Index; CRP, C-reactive protein; COPD, chronic obstructive pulmonary disease; TNF, tumour necrosis factor.

response to HBV vaccine in IBD patients. Indeed, the small subset of patients in this cohort who demonstrated high disease activity scores [CDAI > 150 or Mayo clinic score > 2], achieved significantly lower AIR and EIR rates compared with those who were in remission, in both treatment arms.

The optimal time point for HBV vaccination in the IBD population is still debatable. Current guidelines recommend employment of a 'screen and vaccinate' strategy against HBV infection, as part of the initial evaluation of newly diagnosed IBD patients. This approach is based on the assumption that immune protection against the virus should be obtained before drug-mediated immunosuppression develops. However, current findings from this study suggest that disease remission maintained through immunosuppressive medications [especially purine analogue monotherapy] has an overall beneficial effect on the immune response to hepatitis B vaccine. This positive effect seems to outweigh the negative impact on immune response to the vaccine, inherent to immunosuppressive therapy. Although our findings need further validation in larger cohorts, at the current state of knowledge, physicians caring for IBD patients who achieve remission with immunosuppressant regimens can safely offer them vaccination against HBV.

We have observed an interaction between the vaccine's efficacy and TNF-alpha inhibitor treatment; the rates of successful vaccination were especially low in the Sci-Vac group patients treated with TNF-alpha inhibitors. TNF-alpha has a pivotal role in the innate and adaptive systems' immune response against hepatitis B.²⁰ Consequently, use of TNF-alpha inhibitors has been associated with a growing number of HBV reactivation episodes, presumably related to drug interference with innate and adaptive immune system control of hepatitis B viral replication.²¹ In addition, response to HBV vaccination seems also to be affected by TNF-alpha inhibition. Previous studies of HBV vaccination in IBD patients have shown that treatment with TNF-alpha inhibitors is associated with a poorer primary response

to HBV vaccine¹⁴ as well as with decline in protective anti-HBs titres 12 months following initial vaccination.^{22,23} In our cohort, patients treated with this class of medications showed lower response rates relative to patients treated with immunomodulator therapy [purine analogues]. However, only in the Sci-B-Vac arm did this difference become statistically significant. The reason for this diminished effect, mainly observed in a vaccine with potentially higher immunogenicity, remains unclear and should be further investigated in future studies.

The addition of pre-S2 and especially pre-S1 epitopes used in third-generation vaccines has been shown to elicit potent cellular and humoral immune responses against the viral particles.^{24,25} This presumably accounts for the improved response rates to vaccination with Sci-B-Vac observed in healthy non-responders to second-generation recombinant vaccines, as well as in specialised groups such as end-stage kidney disease and post-liver transplant patients.^{16,17,26,27} However, in our trial, Sci-B-Vac did not confer a similar advantage over the standard HBV vaccine. Although larger-scale trials are needed to validate this finding, the results of this study do not support the use of Sci-B-Vac as an alternative to ENGERIX-B for vaccination against HBV in IBD patients treated with immunosuppressants.

Our study has several limitations. First, only 72 patients were enrolled [we have not reached the proposed sample size due to the limited enrolment], a relatively small number of participants compared with other vaccination studies. This may have had an impact on the primary and secondary outcomes of this study. Due to this limitation, we have calculated conditional power to reject H_0 in a case in which we had recruited 50 subjects per arm as originally planned. The conditional power was 30%. Thus we may conclude that even if we had followed the original recruitment plan, the sample size was too low to reject H_0 . Furthermore, based on the data obtained from the current study, we have calculated the sample size needed to show significant difference between the two vaccines [Sci-B-Vac vs ENGERIX B] above observed ratio 1.18, which showed

that the sample size needs to be 180 subjects per arm. Second, the relatively small number of patients having active disease and/or treated with TNF- α inhibitors in each arm did not allow us to interrogate the individual effect of each of these factors on response to the different vaccines. Third, clinically based disease activity scores and CRP levels were used to determine disease severity. Their use is commonly implemented in interventional studies assessing treatment efficacy in the IBD population, but they do not truly reflect mucosal inflammation as captured by endoscopy or imaging modalities. Finally, although patients in this study were randomised and blinded to treatment assignment, physicians administering the vaccines were not. We cannot exclude that this may have affected the results of this study.

In conclusion, this randomised controlled trial demonstrated no advantage of third-generation multi-epitope vaccine over commonly used second-generation HBV vaccine in patients with IBD treated with immunosuppressant and biological therapy. As in other vaccine studies conducted in this population, the use of TNF- α inhibitors was shown to have a detrimental effect on response to vaccination. The growing use of TNF- α inhibitors in patients with IBD mandates development of alternative approaches aimed at improving vaccination efficacy and long-term protection against HBV in this population.

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Conflict of Interest

None.

Author Contributions

All authors have made substantial contribution to this manuscript as listed below: OE, concept and design of the study, data acquisition and interpretation, drafting of the manuscript and final approval of the version to be submitted; VN, concept and design of the study, analysis and interpretation of data, and critical review of the manuscript; YP, acquisition, analysis, and interpretation of data; OA, data acquisition and drafting of the manuscript; DS, acquisition, analysis, and interpretation of data and critical review of the manuscript; DM, acquisition of data and critical review of the manuscript; NA, acquisition of data and critical review of the manuscript; GB-Y, acquisition of data and critical review of the manuscript; EDM, acquisition, analysis, and interpretation of data; AM, acquisition of data and critical review of the manuscript; VD, acquisition of data and critical review of the manuscript; AF-C, review of the manuscript and final approval of the version to be submitted.

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