Rituximab increases the risk of hepatitis B virus reactivation in non-Hodgkin lymphoma patients who are hepatitis B surface antigen-positive or have resolved hepatitis B virus infection in a real-world setting

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Background. Hepatitis B virus (HBV) reactivation with a hepatitis flare is a common complication in lymphoma patients treated with immunotherapy and/or chemotherapy. Anti-HBV prophylaxis is suggested for non-Hodgkin lymphoma (NHL) patients undergoing rituximab therapy, even those with resolved HBV infection. Since anti-HBV prophylaxis for patients with resolved HBV infection is not covered by national health insurance in Taiwan, a proportion of these patients receive no prophylaxis. In addition, late HBV reactivation has emerged as a new issue in recent reports, and no consensus has been reached for the optimal duration of anti-viral prophylaxis. Thus, the aim of our study was to investigate the incidence and outcomes of HBV reactivation in NHL patients in a real-world setting and to study the frequency of late HBV reactivation.

Materials. NHL patients who received rituximab and/or chemotherapy at our institute between January 2011 and December 2015 and who were hepatitis B surface antigen (HBsAg)- or hepatitis B core antibody (HBcAb)-positive were reviewed retrospectively.

Results. A total of 388 patients were screened between January 2011 and December 2015. In total, 196 patients were excluded because HBsAg was not assessed, HBcAb was negative or not assessed, or they were not treated with immunosuppressive therapy. Finally, the retrospective study included 62 HBsAg-positive NHL patients and 130 NHL patients with resolved HBV infection(HBsAg-negative and HBcAb-positive). During a median 30.5-month follow-up period, seven patients experienced HBV reactivation, five of whom had a hepatitis flare. The incidence of HBV reactivation did not significantly differ between the HBsAg-positive patients and the resolved HBV infection population without anti-HBV prophylaxis(4.8% vs. 3.1%, P=0.683). All patients with HBV reactivation were exposed to rituximab. Notably, late HBV reactivation was not uncommon (two of seven patients with HBV reactivation events, 28.6%). HBV reactivation did not influence the patients' overall survival. An age ≥65 years and an advanced disease stage were independent risk factors for poorer overall survival.

Conclusion. The incidence of HBV reactivation was similar between the HBsAg-positive patients with anti-viral prophylaxis and the resolved HBV infection population without anti-HBV prophylaxis. All HBV reactivation events occurred in NHL patients exposed to rituximab. Late reactivation was not uncommon.

The duration of regular liver function monitoring for more than one year after immunosuppressive therapy or after withdrawal of prophylactic antiviral therapy should be prolonged. Determining the exact optimal duration of anti-HBV prophylaxis is warranted in a future prospective study for NHL patients treated with rituximab containing therapy.

1 **Rituximab increases the risk of hepatitis B virus reactivation in non-Hodgkin lymphoma**
2 **patients who are hepatitis B surface antigen-positive or have resolved hepatitis B virus**
3 **infection in a real-world setting**
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30 **Abstract**

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32 complication in lymphoma patients treated with immunotherapy and/or chemotherapy. Anti-

33 HBV prophylaxis is suggested for non-Hodgkin lymphoma (NHL) patients undergoing rituximab

34 therapy, even those with resolved HBV infection. Since anti-HBV prophylaxis for patients with

35 resolved HBV infection is not covered by national health insurance in Taiwan, a proportion of

36 these patients receive no prophylaxis. In addition, late HBV reactivation has emerged as a new

37 issue in recent reports, and no consensus has been reached for the optimal duration of anti-viral

38 prophylaxis. Thus, the aim of our study was to investigate the incidence and outcomes of HBV

39 reactivation in NHL patients in a real-world setting and to study the frequency of late HBV

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42 **Materials**. NHL patients who received rituximab and/or chemotherapy at our institute between

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46 **Results.** A total of 388 patients were screened between January 2011 and December 2015. In

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48 assessed, or they were not treated with immunosuppressive therapy. Finally, the retrospective

49 study included 62HBsAg-positive NHL patients and 130 NHL patients with resolved HBV

50 infection(HBsAg-negative and HBcAb-positive). During a median 30.5-month follow-up period,

51 seven patients experienced HBV reactivation, five of whom had a hepatitis flare. The incidence

52 of HBV reactivation did not significantly differ between the HBsAg-positive patients and the

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Conclusion

The incidence of HBV reactivation was similar between the HBsAg-positive patients with anti-viral prophylaxis and the resolved HBV infection population without anti-HBV prophylaxis. All HBV reactivation events occurred in NHL patients exposed to rituximab. Late reactivation was not uncommon. The duration of regular liver function monitoring for more than one year after immunosuppressive therapy or after withdrawal of prophylactic antiviral therapy should be prolonged. Determining the exact optimal duration of anti-HBV prophylaxis is warranted in a future prospective study for NHL patients treated with rituximab containing therapy.

Introduction

Hepatitis B virus (HBV) infection is a primary cause of chronic liver disease, liver cirrhosis, and hepatocellular carcinoma. Approximately 15–20% of adults in Taiwan are chronically infected with HBV(Sung,1984;Gust, 1996), and chronic liver disease and cirrhosis was the tenth leading cause of death in 2017. Therefore, Taiwan is a hyperendemic area for HBV infection, and HBV infection is an important health issue in Taiwan.

According to the guideline from the American Association for the Study of Liver Disease (AASLD), HBV reactivation in hepatitis B surface antigen (HBsAg)-positive patients is defined as a ≥2 log increase in HBV DNA compared to baseline or an HBV DNA level ≥1000 IU/mL in patients with previously undetectable levels. HBV reactivation in patients with resolved HBV infections is defined as the reappearance of HBsAg or detectable HBV DNA. A hepatitis flare is defined as an increase in alanine aminotranferase (ALT) to greater than or equal to three times the baseline level and>100 IU/L (Terrault et al., 2018).

Reports of HBV reactivation after rituximab therapy started emerging a few years after FDA approval.HBV reactivation and hepatitis flares are well known complications in cancer patients receiving chemotherapy. The combination regimen of rituximab and cytotoxic chemotherapy has also been found to increase the risk of hepatitis B reactivation, even in patients in whom HBV infection is resolved (i.e., HBsAg-negative but hepatitis B core antibody [HBcAb]-positive)(Dong et al., 2013; Evens et al., 2011; Yeo et al., 2009).Due to the high prevalence rate of chronic HBV infection in Taiwan, HBV reactivation-related complications are a great concern in lymphoma patients receiving rituximab and chemotherapy.

The recently published guidelines from the European Association for the Study of the Liver

90   (EASL) and the AASLD both recommend that patients undergoing rituximab therapy receive

91   anti-HBV prophylaxis(Lampertico et al., 2017; Terrault et al., 2018). However, the

92   recommended duration of anti-HBV prophylaxis is not consistent. Whereas the AASLD suggests

93   that anti-HBV prophylaxis should continue for at least six months (or for at least 12 months for

94   patients receiving rituximab) after the completion of immunosuppressive therapy, the EASL

95   suggests that prophylaxis should continue for at least 12 months (or for at least 18 months for

96   rituximab-based therapy). Moreover, late HBV reactivation (i.e., reactivation more than 12

97   months after completing immunosuppressive therapy(Nakaya et al., 2016) or after withdrawal of

98   prophylactic antiviral therapy(Liu et al., 2016)) has been reported recently. However, the optimal

99   duration of anti-HBV prophylaxis has not been determined. Furthermore, because anti-HBV

100   prophylaxis for patients with resolved HBV infection is not covered by national health insurance

101   in Taiwan, prophylactic anti-HBV therapy is not prescribed routinely to these patients.

102   Therefore, we performed a retrospective study to understand the reality of HBV reactivation in

103   patients with resolved HBV infection and to investigate the incidence and outcomes of HBV

104   reactivation and late HBV reactivation in HBsAg-positive NHL patients and in NHL patients

105   with resolved HBV infections who were being treated with rituximab and/or chemotherapy.

106

107   **Materials & Methods**

108   **Patients and study design**

109   We performed a retrospective observational study of patients newly diagnosed with NHL at

110   Kaohsiung Medical University Hospital between January 2011 and December 2015. The

111   inclusion criteria were as follows: 1) age 20 years or older, 2) received at least one cycle of

112   rituximab and/or chemotherapy, and 3) were initially either HBsAg- or HBcAb-positive. The

113   collected data included demographics, histological subtypes, treatment choice, hepatitis virus

114   serology, HBV DNA level, and patient outcome. The baseline hepatitis virus serology and the

115   ALT and HBV DNA level were tested during the time interval from the diagnosis of lymphoma

116   to the first treatment. HBsAg-positive patients took their anti-viral prophylaxis until six months

117   after cessation of chemotherapy, which was covered by national health insurance. HBsAg-

118   positive patients received regular follow-up evaluations of their hepatitis virus serology and

119   HBV DNA levels every three months, whereas patients with resolved HBV infection only

120   received hepatitis virus serological testing when the ALT level became abnormal. Overall

121   survival was defined as the length of time from the date of diagnosis to the last date of follow-up

122   or death. All patients were followed up until February 2017.

123   Informed consent was not needed because the study was a retrospective chart review. The study

124   was approved by the Institutional Review Board of Kaohsiung Medical University Hospital,

125   Kaohsiung, Taiwan, in 2016 (*KMUHIRB-E(I)-20160009*).

126

127   **Statistical analysis**

128   Patient characteristics and factors contributing to HBV reactivation were analyzed using

129   descriptive statistics and are presented as frequencies, percentages, and medians. Student's t-test

130   or nonparametric statistics were utilized to test for statistically significant differences in

131   continuous variables, whereas the chi-square or Fisher's exact test was used for categorical

132   variables. Overall survival was estimated using the Kaplan-Meier method. Variables that were

133   statistically significant (*P*<0.05) in the univariate analysis of survival were subsequently

134   subjected to multivariate analysis using a Cox regression model. A two-tailed *P*-value <0.05 was

135   considered statistically significant.

136

137   **Results**

138   **Patient characteristics**

139   A total of 388 patients were diagnosed with NHL between January 2011 and December

140   2015. Seventy patients were positive for HBsAg, and 310 were negative. Among the HBsAg-

141   negative patients, 158 were positive for HBcAb, 130 of whom received at least one cycle of

142   rituximab or chemotherapy and thus were included in the study. Among the HBsAg-positive

143   patients, 62 received at least one cycle of rituximab or chemotherapy and were also included. A

144   flowchart of the patient selection process is shown in Figure 1.

145   Among all included patients, 54.7% were male, and the median age at the time of diagnosis

146   was 62 years (range, 27–90 years). The median length of follow-up was 30.5 months (range 0.8-

147   73.9 months). The baseline patient characteristics are summarized in Table 1. The HBsAg-

148   positive group exhibited a lower proportion of patients who were positive for HBsAb than the

149   HBsAg-negative/HBcAb-positive group (i.e., the resolved HBV infection) (14.5% vs. 72.3%,

150   *P*<0.001) and was the only group to receive prophylactic anti-viral therapy (98.4% vs. 0%,

151   *P*<0.001). No differences were found in the age, sex, lymphoma subtype, stage, rituximab use,

152   and HBV reactivation rate between these two groups.

153

154   **Incidence of HBV reactivation and HBV-related hepatitis flares**

155   Overall, seven patients developed HBV reactivation with an incidence rate of 3.6%, and

156   five patients experienced an HBV-related hepatitis flare. Three of the seven patients with HBV

157   reactivation were HBsAg-positive, and four had a resolved HBV infection at the initial

158   diagnosis. The incidence rates of HBV reactivation in the HBsAg-positive and resolved HBV

159   infection groups were 4.8% and 3.1%, respectively, which were not significantly different.

160

161   **Factors associated with HBV reactivation**

162   The factors attributed to HBV reactivation are compared in Table 2. Most patients

163   experiencing HBV reactivation were older (≥60 years), had advanced-stage disease, and were

164   predominantly male. All patients who experienced HBV reactivation received rituximab-

165   containing treatment.

166

167   **HBV reactivation and HBV-related hepatitis flares in the HBsAg-positive patients**

168   Three of the 62 HBsAg-positive patients developed HBV reactivation. The details for these

169   three patients are shown in Table 3. Two of these patients were male. All patients were in the

170   advanced stage four, received multiple cycles of rituximab-containing treatment, and were

171   receiving a different antiviral agent than that prescribed for their previous prophylaxis. Two

172   patients developed a hepatitis flare. Patient number one received entecavir for 7.9 months, and

173   their hepatitis was resolved without a detectable HBV DNA level. Patient number three received

174   both tenofovir and entecavir but still died of fatal fulminant hepatitis attributed to HBV

175   reactivation (although this patient also had comorbid hepatocellular carcinoma).

176

177   **Comparison of HBV reactivation rates between subgroups divided based on antiviral**

178   **prophylaxis**

179   Sixty-one of the 62 (98.4%) HBsAg-positive patients received anti-HBV prophylaxis. The

180   anti-HBV prophylactic drugs administered to the HBsAg-positive patients included entecavir (35

181   of 61, 57.4%), tenofovir (13 of 61, 21.3%), lamivudine (nineof 61, 14.7%), and telbivudine (four

182   of 61, 6.6%). A higher HBV reactivation rate (one of nine, 11.1%) was found in the patients who

183   used lamivudine as antiviral prophylaxis than in those who used entecavir, tenovir, or telbivudine

184   (two of 52, 3.8%).

185

186   **HBV reactivation and HBV-related hepatitis flares in the patients with resolved HBV**

187   **infections**

188   Four of 130 patients with resolved HBV developed HBV reactivation; these patients are

189   presented in Table 4.Three patients were diagnosed with a hepatitis flare, all of whom recovered

190   after antiviral treatment (only one used lamivudine alone, whereas the other three patients used

191   tenofovir or entecavir). Among these four HBV reactivated patients, three were male and had

192   stage four disease. All patients were exposed to multiple cycles of rituximab-containing

193   chemotherapy. One patient died of lymphoma progression not associated with a hepatitis flare.

194

195   **Late HBV reactivation**

196   Two patients experienced late HBV reactivation. The probability of late HBV reactivation

197   was not uncommon in the patients who experienced a reactivation event (28.6%). One patient

198   had HBV that was initially resolved but reactivated 36.3 months after completing R-CHOP

199   (rituximab with cyclophosphamide, doxorubicin, vincristine, and prednisolone) treatment; the

200   other was initially HBsAg-positive and experienced HBV reactivation 11.2 months after

201   withdrawal of prophylactic antiviral therapy.

202

203   **Overall survival of NHL patients**

204   The five-year overall survival rate was 71.0% based on the Kaplan-Meier analysis. The

205   survival plot is shown in Figure 2. We ran a Cox regression model to analyze factors influencing

206   the patients' mortality (Table 5). The variables with a significant effect on mortality in the

207   univariate analysis were included in the multivariate analysis. An older age (hazard ratio [HR]

208   1.03, 95% confidence interval [CI] 1.01–1.06, P=0.005) and advanced disease stage (HR 4.21,

209   95% CI 1.78–9.95, P=0.001) were associated with inferior survival, whereas the HBsAg status,

210   HBV reactivation, and hepatitis flare did not influence overall survival.

Discussion

211

212   Our study provided real-world data for the incidence and outcomes of HBV reactivation and

213   hepatitis flares in NHL patients. We found that the overall incidence of HBV reactivation was

214   3.6%; the incidence rates of HBV reactivation in the HBsAg-positive group (receiving

215   prophylaxis) and the resolved HBV infection group (which did not receive prophylaxis)

216   were4.8% and 3.1%, respectively, which were not significantly different.

217   A retrospective Asia Lymphoma Study Group (ALSG) study of HBV reactivation in

218   lymphoma patients revealed that this event occurred in 27.8% of HBsAg-positive patients and

219   was significantly less frequent in patients receiving antiviral prophylaxis than in those not

220   receiving it (22.9% vs. 59.1%; *P*<0.001)(Kim et al., 2013).By comparison, we observed a

221   significantly lower HBV reactivation rate (only 4.8%) in our HBsAg-positive patients, 98.4% of

222   whom received prophylactic anti-viral therapy. Lamivudine has proven to be a useful drug to

223   prevent HBV reactivation in patients undergoing chemotherapy for NHL(Persico et al., 2002).

224   However, several studies demonstrated that patients receiving second-line entecavir as a

225   prophylactic agent exhibited a significantly lower HBV reactivation rate than those receiving

226   lamivudine(Chen et al., 2013; Chen et al., 2015; Huang et al., 2014; Li et al., 2011).Furthermore,

227   the AASLD 2018 hepatitis B guidelines recommended entecavir and tenofovir as the preferred

228   first-line anti-HBV agents because of their higher potency and barrier to resistance (Terrault et

229   al., 2018). Notably, most patients in the ALSG study used lamivudine, whereas more than 75%

230   of the patients in our study were treated with entecavir and tenofovir for HBV prophylaxis. In

231   addition, a higher HBV reactivation rate (11.1%) was noted in patients who used lamivudine as

232   antiviral prophylaxis than in those who used entecavir, tenovir, or telbivudine (3.8%).This result

233   may explain the lower HBV reactivation rate (4.8%) in our HBsAg-positive patients.

234   Among the population with resolved HBV infections in the ALSG study, 9.6% developed

235   HBV reactivation (Kim et al., 2013), whereas the HBV reactivation rate in our study was 3.1%.

236   Several studies have found that HBsAb negativity or a low titer is a risk factor for HBV

237   reactivation (Cho et al., 2016; Matsubara et al., 2017; Seto et al., 2014; Yeo et al., 2009).The

238   higher percentage (72.3%) of HBsAb-positive patients in our resolved HBV infection group than

239   in our HBsAg-positive group might partially explain why these patients did not experience a

240   higher HBV reactivation rate even in the absence of anti-HBV prophylaxis. Another key factor

241   was that we did not routinely monitor the HBV DNA and ALT levels in the patients with

242   resolved HBV infections and only tested their HBV status upon detecting abnormal liver

243   function. This strategy may have led to underestimation of the incidence of HBV reactivation in

244   our resolved HBV infection group.

245   Regarding the features of the patients who experienced HBV reactivation, receiving a

246   rituximab-containing regimen, an older age (≥60 years), male sex, and an advanced disease stage

247   were associated with HBV reactivation. All patients who experienced HBV reactivation received

248   rituximab for at least five cycles; this finding was consistent with a recent meta-analysis that

249   revealed a significantly higher rate of HBV reactivation (8.2%) in patients receiving rituximab-

250   containing chemotherapy than in those not receiving rituximab (0.6%)(Evens et al., 2011). In

251   addition, a study focused on elderly (age>65 years) NHL patients with resolved HBV infections

252   also reported that HBV reactivation was more likely to occur after an average of five R-

253   CHOP cycles(Castelli et al., 2016). These findings all implied that multiple cycles of rituximab-

254   containing chemotherapy was the key factor for HBV reactivation.

255   One patient died due to HBV reactivation-induced fatal fulminant hepatitis. However, the

256   patient also had an underlying hepatocellular carcinoma malignancy. The remaining four

257   hepatitis flare patients resolved after antiviral treatment, and three achieved an undetectable

258   HBV DNA level. Overall, HBV reactivation and hepatitis flares did not compromise patient

259   survival in our study. This result was consistent with a previous study conducted in north Taiwan

260   in which HBV reactivation was associated with a greater number of cycles (greater than or equal

261   to six) and a prolonged duration of rituximab therapy; overall survival in that study did not differ

262   between patients with versus those without HBV reactivation (Hsiao et al., 2015). In our study,

263   an age ≥60 years and Ann Arbor stages III–IV were two independent negative prognostic factors.

264   The EASL and AASLD guidelines both recommend that patients who undergo rituximab

265   therapy should receive anti-HBV prophylaxis(Lampertico et al., 2017; Terrault et al., 2018);

266   however, the optimal duration of such prophylaxis remains uncertain and has varied from six to

267   18 months after completion of immunosuppressive therapy. A retrospective study conducted by

268   Liu et al. (2016) found that the incidence rate of HBV reactivation after withdrawal of

269   prophylactic antiviral therapy was 21.7% and that the median time from the withdrawal of the

270   prophylactic antiviral therapy to HBV reactivation was 2.9 months (range, 1.1–8.5)(Liu et al.,

271   2016).However, several studies also reported that patients developed late HBV reactivation (i.e.,

272   more than one year post-therapy)(Lee et al., 2010; Muraishi et al., 2017; Nakaya et al., 2016;

273   Yamada et al., 2017). The literature review of Yamada et al.(2017) revealed that an advanced

274   disease stage, lymphoid malignancies, and treatment with multiple courses of rituximab-

275   containing therapies were associated with late HBV reactivation (Yamada et al., 2017).In our

276   study, late HBV reactivation accounted for 28.6% of the total HBV reactivation events and thus

277   was not as rare as we expected. The longest time to HBV reactivation after completion of

278   rituximab-containing therapy was 36.3 months. Hence, these data indicate that close monitoring

279   of the HBV DNA and ALT levels remains important even at one year after completing

280   rituximab-containing therapy or withdrawal of prophylactic anti-viral therapy. Identifying

281   patients at a high risk of late HBV reactivation and determining the optimal duration of

282   prophylaxis for high-risk populations are pivotal aims that require further study.

283   Although we obtained notable results, our study had some limitations. First, our study was a

284   retrospective observational study with a small number of patients in each group. Second, the

285   differing chemotherapy regimens and doses may have constituted confounding factors in terms

286   of HBV reactivation. Third, the resolved HBV infection group did not receive routine follow-up

287   (including liver function and HBV statuses). However, our study provides clinicians the real-

288   world data for HBV reactivation and reminds them of the possibility of late HBV reactivation.

289

290   **Conclusions**

291   The incidence of HBV reactivation is similar between HBsAg-positive patients with anti-

292   viral prophylaxis and patients with resolved HBV infections without anti-HBV prophylaxis.

293   Rituximab exposure is a key risk factor for HBV reactivation. Late HBV reactivation is not as

294   rare as we expected, and regular monitoring of the HBV DNA and ALT levels is important even

295   one year after withdrawal of immunosuppressive therapy or prophylactic anti-viral therapy.

296   Prospective studies are required to determine the optimal duration of prophylaxis for patients at

297   high risk of late HBV reactivation.

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301   **References**

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Figure 1

Flow chart of patient selection

Abbreviations: HBsAg, hepatitis B surface antigen; HBcAb, hepatitis B core antibody; *H. pylori, Helicobacter pylori*

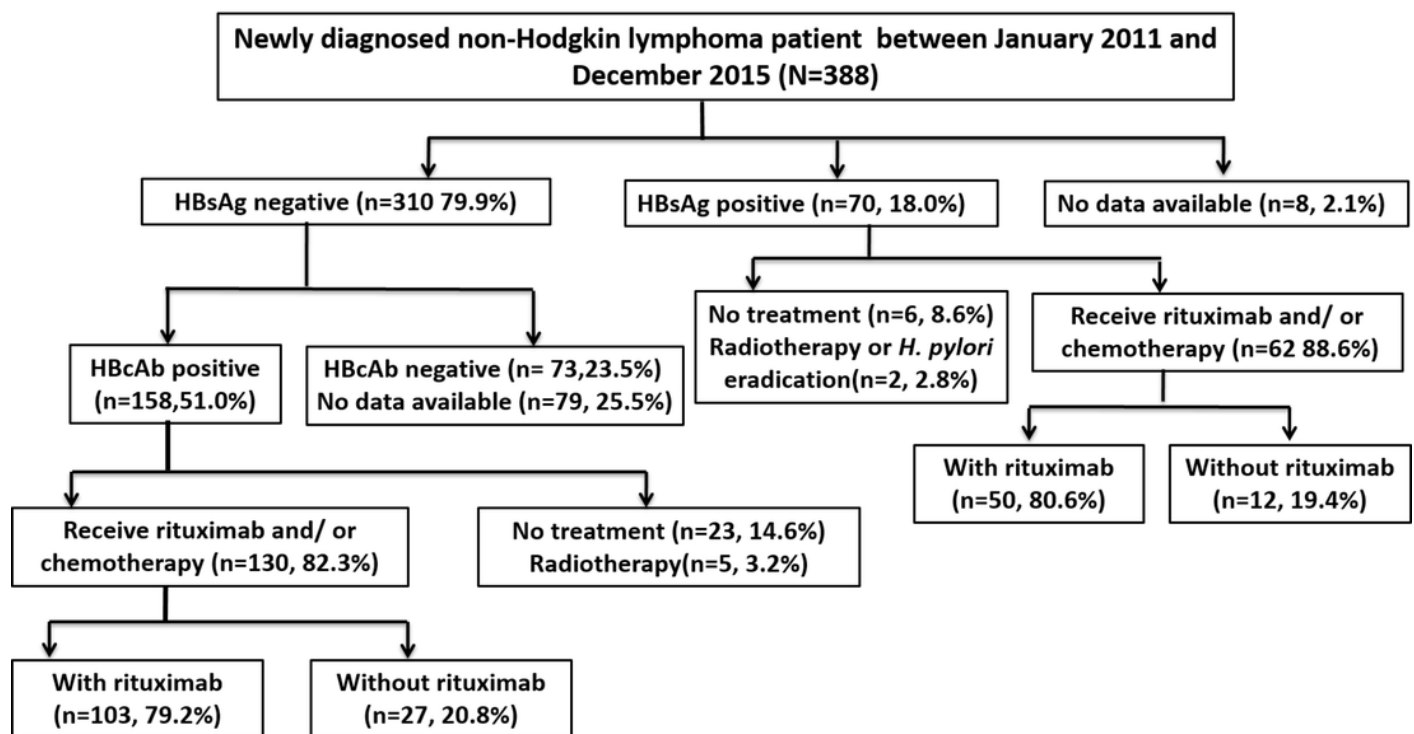



Figure 2

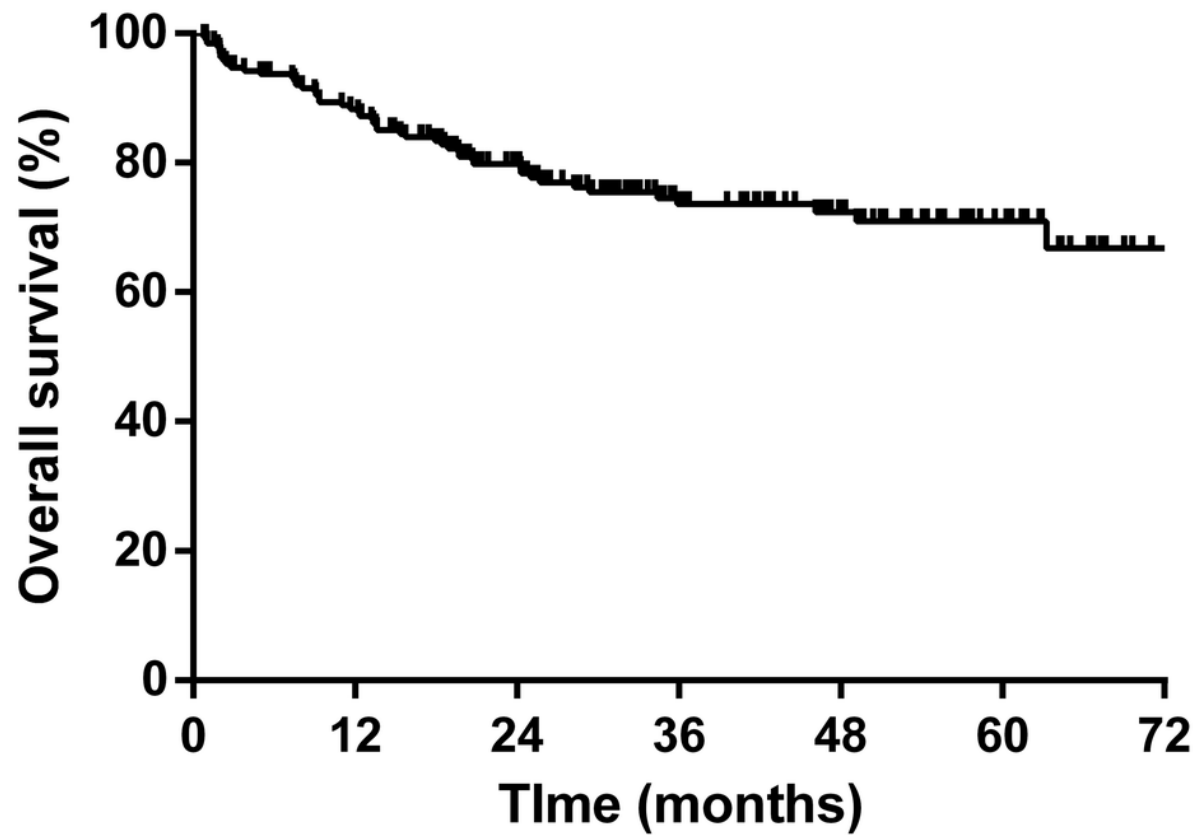
Kaplan-Meier survival curve of overall survival for all patients

Baseline characteristics in non-Hodgkin lymphoma patients who received rituximab and/or chemotherapy

Table 1. Baseline characteristics in non-Hodgkin lymphoma patients who received rituximab and/or chemotherapy

	All patients	HBsAg-positive group	HBsAg-negative/HBcAb-positive	*P*-value
Sex				0.977
Male	105(54.7)	34(54.8)	71(54.6)	
Female	87(45.3)	28(45.2)	59(45.4)	
Age (years)(median, range)	62 (27–90)	56(27–83)	63(28–90)	0.885
HBsAb				0.000
Positive	103(53.6)	9(14.5)	94 (72.3)	
Negative	75(39.1)	49(79.0)	26 (20.0)	
Data unavailable	14(7.3)	4(6.5)	10(7.7)	
Lymphoma subtype				0.494
DLBCL	118(61.5)	36(58.1)	82(63.1)	
FL	19(9.9)	8(12.9)	11(8.5)	
Marginal zone lymphoma	21(10.9)	7(11.3)	14(10.8)	
T cell lymphoma	10(5.2)	1(1.6)	9(6.9)	
MALToma	9(4.7)	4(6.5)	5(3.8)	
Mantle cell lymphoma	7(3.6)	2(3.2)	5(3.8)	
Burkitt lymphoma	1(0.5)	0	1(0.8)	
Other B cell lymphoma[a]	7(3.6)	4(6.5)	3(2.3)	
Stage				0.194
I	31(16.1)	13(21.0)	18(13.8)	
II	32(16.7)	6(9.7)	26(20.0)	
III	42(21.9)	12(19.4)	30(23.1)	
IV	87(45.3)	31(50.0)	56(43.1)	
With rituximab therapy	153(79.7)	50(80.6)	103(79.2)	0.820
Prophylaxis with anti-HBV agents	61(31.8)	61(98.4)	0(0)	0.000
HBV reactivation	7(3.6)	3(4.8)	4(3.1)	0.683

1   [a]Other B cell lymphomas: 1 B cell lymphoma, 1 large B cell lymphoma, 1 B-cell lymphoma, unclassifiable with features intermediate between diffuse large B-

2   cell lymphoma and classical Burkitt's lymphoma, 1 lymphoplasmacytic lymphoma, 1 plasmablastic lymphoma, and 2 small lymphocytic lymphoma.

3   Abbreviations: DLBCL, diffuse large B cell lymphoma; FL, follicular lymphoma; MALT, mucosa associated lymphoid tissue; HBV, hepatitis B virus; HBsAg,

4   hepatitis B surface antigen; HBcAb, hepatitis B core antibody; HBsAb, hepatitis B surface antibody.

Factors influencing HBV reactivation in NHL patients under rituximab and/or chemotherapy treatment

1 **Table 2. Factors influencing HBV reactivation in NHL patients under rituximab and/or chemotherapy treatment**

Factors	All patients				HBsAg positive				HBsAg negative/HBcAb positive			
	HBV reactivation		No HBV reactivation		HBV reactivation		No HBV reactivation		HBV reactivation		No HBV reactivation	
	N=7	%	N=185	%	N =3	%	N=59	%	N =4	%	N=126	%
Age												
<60	2	28.6	82	44.3	1	33.3	35	59.3	1	25.0	47	37.3
≥60	5	71.4	103	55.7	2	66.7	24	40.7	3	75.0	79	62.7
Sex												
Male	5	71.4	100	54.1	2	66.7	32	54.2	3	75.0	68	54.0
Female	2	28.6	85	45.9	1	33.3	27	45.8	1	25.0	58	46.0
Ann Arbor stage												
I–II	1	14.3	62	33.5	0	0	19	32.2	1	25.0	43	34.1
III–IV	6	85.7	123	66.5	3	100	40	67.8	3	75.0	83	65.9
HBsAb												
-	2	28.6	73	39.5	1	33.3	48	81.4	1	25.0	25	19.8
+	4	57.1	99	53.5	1	33.3	8	13.5	3	75.0	91	72.2
Unavailable	1	14.3	13	7.0	1	33.3	3	5.1	-	-	10	7.9
Rituximab												
-	0	0	39	21.1	0	0	12	20.3	0	0	27	21.4
+	7	100	146	78.9	3	100	47	79.7	4	100	99	78.6

2   Abbreviations: HBV, hepatitis B virus; NHL, non-Hodgkin lymphoma; HBsAg, hepatitis B surface antigen; HBcAb, hepatitis B core antibody; HBsAb, hepatitis

3   B surface antibody

Details of HBV reactivation and HBV-related hepatitis flares in the HBsAg-positive patients

1   **Table 3. Details of HBV reactivation and HBV-related hepatitis flares in the HBsAg-positive patients**

Patient number	Sex	Age	diagnosis	Stage	HBsAb	Baseline HBVDNA level (KIU/ml)	Prophylaxis Anti-HBV agents	Treatment regimen (# of courses)	Time to HBV reactivation (months)	HBV DNA level when reactivation (KIU/ml)	Hepatitis flare (ALT level IU/L)	Outcome and anti-viral treatment after HBV reactivation	Survival state
1	Male	62	FL	4	Unknown	0.727	Telbivudine	R-CHOP (8)	11.2 months after withdrawal of prophylaxis (late reactivation)	44700	Yes (137)	ALT normal, HBV DNA level undetectable after 7.9 months of entecavir	Alive
2	Female	37	FL	4	Negative	5.04	Entecavir	R-CHOP (8) with R maintain (8)	During chemotherapy, 12.7 months after the first time of chemotherapy	92200	No (28)	Last HBV DNA level 0.618 (KIU/ml) after 18.6 months of tenofovir	Alive
3	Male	67	DLBCL (HCC)	4	Positive	27.8	Tenofovir for 2 years then switched to Lamivudine	RCHOP (5) RESHAP (4)	15.4 months after the last time of chemotherapy but still on lamivudine	85900	Yes (2050)	Tenofovir and entecavir for one week and die due to hepatic failure	Died of hepatic failure

								prophylaxis					

2   Abbreviations: ALT,alanine transaminase; DLBCL, diffuse large B cell lymphoma; FL, follicular lymphoma; HCC, hepatocellular carcinoma; R-CHOP,

3   rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisolone; R-ESHAP, rituximab, etoposide, solu-medrol, cytarabine, and cisplatin; HBsAg,

4   hepatitis B surface antigen; HBsAb, hepatitis B surface; R maintain, Rituximab maintenance.

Details of HBV reactivation and HBV-related hepatitis flares in the patients with resolved HBV infections

1   **Table 4. Details of HBV reactivation and HBV-related hepatitis flares in the patients with resolved HBV infections**

Patient number	Sex	Age (years)	diagnosis	Stage	Anti-HBs	Treatment regimen (# of cycles)	HBV reactivation time	HBV DNA (KIU/ml)	Hepatitis flare (ALT level: IU/L)	Outcome and anti-viral treatment after HBV reactivation	Survival state
1	Female	68	FL	4	Negative	RCHOP (8)	36.3 months after last course of R-CHOP (late reactivation)	2.52*	Yes (537)	ALT normal, HBV DNA level undetectable after 13.9 months of lamivudine	Alive
2	Male	50	Marginal zone B cell lymphoma and transform to DLBCL	4	Positive	CHOP (8), ESHAP (6), R-ICE (5), auto-HSCT EPOCH (1)	During treatment, 56.0 months after 1st course of chemotherapy	124000	Yes (121)	ALT normal, last HBV DNA level: 0.445 KIU/ml after 4.3 months of tenofovir	Died of lymphoma progression
3	Male	73	DLBCL	2	Positive	RCHOP (6)	8.4 months after last course of R-CHOP	12200	Yes (863)	ALT normal, HBV DNA level undetectable after entecavir	Alive

										and lamivudine together for 2 weeks and then only entecavir for 3.9 months	
4	Male	60	FL	4	Positive	RCHOP (5) followed by R maintain(8)	During treatment, 24.5 months after 1st course of chemotherapy,	>170000	No (98)	Just start tenofovir treatment and still pending the HBV DNA level data	Alive

2 Abbreviations: ALT, alanine transaminase; DLBCL, diffuse large B cell lymphoma; EPOCH, etoposide, prednisolone, vincristine, cyclophosphamide, and doxorubicin;ESHAP,

3 etoposide, solu-medrol, cytarabine, and cisplatin; FL, follicular lymphoma; HSCT, hematopoietic stem cell transplantation; (R)-CHOP, (rituximab)-cyclophosphamide,

4 doxorubicin, vincristine, and prednisolone; ESHAP, etoposide, solu-medrol, cytarabine, and cisplatin; R-ICE, rituximab- ifosfamide, carboplatin, etoposide; HBsAg, hepatitis B

5 surface antigen; HBcAb, hepatitis B core antibody; HBsAb, hepatitis B surface antibody; R maintain, Rituximab maintenance.*: HBV viral load data was checked after anti-viral

6 treatment

Cox proportional hazard ratios for mortality in NHL patients

1   Table 5. Cox proportional hazard ratios for mortality in NHL patients

variables	Mortality		Mortality (Adjusted)	
	HR (95%CI)	*P*-value	HR (95%CI)	*P*-value
Gender (male vs. female)	1.59(0.88-2.89)	0.120		
Age	1.03(1.01-1.05)	0.013*	1.03(1.01-1.06)	0.005*
Stage (III and IV vs. I and II)	3.86(1.64-9.09)	0.002*	4.21(1.78-9.95)	0.001*
Rituximab use	0.65(0.34-1.22)	0.179		
HBsAg positive	0.69(0.36-1.32)	0.260		
HBV reactivation	0.82(0.20-3.38)	0.779		
HBV related hepatitis flare	0.92(0.22-3.82)	0.908		

2   Abbreviations: CI: confidence interval, HR: hazard ratio, NHL: non-Hodgkin lymphoma

3   *: p<0.05