ORIGINAL ARTICLE

Bulevirtide Combined with Pegylated Interferon for Chronic Hepatitis D

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ABSTRACT

BACKGROUND

In a phase 3 trial, bulevirtide monotherapy led to a virologic response in patients with chronic hepatitis D. Pegylated interferon (peginterferon) alfa-2a is recommended by guidelines as an off-label treatment for this disease. The role of combination therapy with bulevirtide and peginterferon alfa-2a, particularly with regard to finite treatment, is unclear.

METHODS

In this phase 2b, open-label trial, we randomly assigned patients to receive peginterferon alfa-2a alone (180 μg per week) for 48 weeks; bulevirtide at a daily dose of 2 mg or 10 mg plus peginterferon alfa-2a (180 μg per week) for 48 weeks, followed by the same daily dose of bulevirtide for 48 weeks; or bulevirtide at a daily dose of 10 mg alone for 96 weeks. All the patients were followed for 48 weeks after the end of treatment. The primary end point was an undetectable level of hepatitis D virus (HDV) RNA at 24 weeks after the end of treatment. The primary comparison was between the 10-mg bulevirtide plus peginterferon alfa-2a group and the 10-mg bulevirtide monotherapy group.

RESULTS

A total of 24 patients received peginterferon alfa-2a alone, 50 received 2 mg and 50 received 10 mg of bulevirtide plus peginterferon alfa-2a, and 50 received 10 mg of bulevirtide monotherapy. At 24 weeks after the end of treatment, HDV RNA was undetectable in 17% of the patients in the peginterferon alfa-2a group, in 32% of those in the 2-mg bulevirtide plus peginterferon alfa-2a group, in 46% of those in the 10-mg bulevirtide plus peginterferon alfa-2a group, and in 12% of those in the 10-mg bulevirtide group. For the primary comparison, the between-group difference was 34 percentage points (95% confidence interval, 15 to 50; P<0.001). At 48 weeks after the end of treatment, HDV RNA was undetectable in 25% of the patients in the peginterferon alfa-2a group, in 26% of those in the 2-mg bulevirtide plus peginterferon alfa-2a group, in 46% of those in the 10-mg bulevirtide plus peginterferon alfa-2a group, and in 12% of those in the 10-mg bulevirtide group. The most frequent adverse events were leukopenia, neutropenia, and thrombocytopenia. The majority of adverse events were of grade 1 or 2 in severity.

CONCLUSIONS

The combination of 10-mg bulevirtide plus peginterferon alfa-2a was superior to bulevirtide monotherapy with regard to an undetectable HDV RNA level at 24 weeks after the end of treatment. (Funded by Gilead Sciences; MYR 204 ClinicalTrials.gov number, NCT03852433.)

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Chronic hepatitis D is caused by the hepatitis D virus (HDV), a defective virus that requires hepatitis B surface antigen (HBsAg) for assembly and propagation.[1] HDV infection affects an estimated 10 to 20 million persons worldwide.[1] As the most severe form of chronic viral hepatitis,[2-4] chronic hepatitis D is associated with a risk of hepatocellular carcinoma that is two to six times as high[5,6] and a risk of death that is two to three times as high as the risks associated with hepatitis B virus (HBV) monoinfection.[7,8]

Until recently, off-label therapy with pegylated interferon (peginterferon) alfa was the only treatment for chronic hepatitis D and was recommended by clinical practice guidelines as the standard of care in eligible patients.[9] However, interferons have several contraindications and are associated with early discontinuation due to side effects and with late relapse after treatment cessation.[10,11] The sodium taurocholate cotransporting polypeptide (NTCP) receptor is the entry receptor for HBV and HDV. A phase 3 trial showed that monotherapy with bulevirtide, a new NTCP entry inhibitor, was associated with greater virologic responses than control (no treatment) at week 48, although the percentage of patients with an undetectable HDV RNA level was low.[12] The phase 2 MYR 203 trial, which had only 15 patients in each treatment group, showed that bulevirtide plus peginterferon alfa-2a for 48 weeks was associated with a higher percentage of patients with an undetectable HDV RNA level at 24 weeks after treatment cessation than was monotherapy with either bulevirtide or peginterferon alfa-2a.[13]

Bulevirtide was the first approved treatment for adults with chronic hepatitis D and compensated liver disease in Europe, and no therapies for chronic HDV infection have been approved in the United States. Guidelines from the European Association for the Study of the Liver recommend that treatment with bulevirtide be considered in all patients with chronic hepatitis D and compensated liver disease and that peginterferon alfa-2a given alone or in combination with bulevirtide be considered in eligible patients.[14] However, whether bulevirtide combined with peginterferon alfa-2a would enhance viral suppression such that finite treatment could be possible has been unclear. We undertook a phase 2b trial (MYR 204) to evaluate bulevirtide, with or without peginterferon alfa-2a, for 96 weeks as a potential finite treatment regimen in patients with chronic hepatitis D.

METHODS

PATIENTS

We enrolled patients 18 to 65 years of age who had chronic hepatitis D, with positive HDV RNA detected by polymerase chain reaction, as well as an alanine aminotransferase level of more than 1 time but less than 10 times the upper limit of the normal range at screening. Key exclusion criteria were decompensated cirrhosis, defined as class B or C cirrhosis according to the Child–Turcotte–Pugh classification (classes range from A to C, with higher classes indicating more severely impaired liver function); the receipt of interferon therapy within the 6 months before screening; and a platelet count of less than 90,000 cells per cubic millimeter. Full inclusion and exclusion criteria are provided in the protocol, which is available with the full text of this article at NEJM.org.

TRIAL DESIGN AND OVERSIGHT

In this multicenter, open-label, randomized, controlled, phase 2b trial, eligible patients were assigned in a 1:2:2:2 ratio to one of four treatment groups: subcutaneous peginterferon alfa-2a (180 μg per week) for 48 weeks (peginterferon alfa-2a group); subcutaneous bulevirtide at a dose of 2 mg per day for 96 weeks with subcutaneous peginterferon alfa-2a (180 μg per week) for the first 48 weeks (2-mg bulevirtide plus peginterferon alfa-2a group); subcutaneous bulevirtide at a dose of 10 mg per day for 96 weeks, with subcutaneous peginterferon alfa-2a (180 μg per week) for the first 48 weeks (10-mg bulevirtide plus peginterferon alfa-2a group); or subcutaneous bulevirtide at a dose of 10 mg day for 96 weeks (10-mg bulevirtide monotherapy group). Randomization was stratified according to the presence of liver cirrhosis. All the trial groups were followed for an additional 48 weeks after the end of treatment (Fig. S1 in the Supplementary Appendix, available at NEJM.org).

The trial was conducted in accordance with the principles of the Declaration of Helsinki, the Good Clinical Practice guidelines of the International Council for Harmonisation, and the trial protocol, which was approved by the independent ethics committee of each participating site. The

sponsor, Gilead Sciences, designed and oversaw the data collection and analysis. Professional writers who were paid by the sponsor prepared the first draft of the manuscript under the authors' direction. All the authors vouch for the accuracy and completeness of the data and for fidelity of the trial to the protocol.

END POINTS AND ASSESSMENTS

The primary end point was an undetectable HDV RNA level, which was defined as the level below the lower limit of quantification with HDV RNA not detected at week 24 after the end of treatment. The end of treatment was at week 48 in the peginterferon alfa-2a group and at week 96 in all the groups that received bulevirtide. Secondary efficacy end points included undetectable HDV RNA at week 48 (in all groups) and week 96 (in all bulevirtide groups) during treatment and at week 48 after the end of treatment (in all groups).

The end point of normalization of the alanine aminotransferase level, which was defined as an alanine aminotransferase value within the normal range on the basis of central laboratory findings (i.e., a value of ≤31 U per liter in women and of ≤41 U per liter in men at sites in Russia or a value of ≤34 U per liter in women and of ≤49 U per liter in men at all other sites), and the composite end point of an undetectable HDV RNA level and normalization of the alanine aminotransferase level were measured at weeks 48 and 96 and at 24 weeks and 48 weeks after the end of treatment. HBsAg loss was assessed at 24 weeks and 48 weeks after the end of treatment. Safety assessments included adverse events and elevations in bile acid levels.

HDV RNA quantification levels were assessed with the use of the RoboGene kit, version 2.0 (lower limit of quantification, 50 IU per milliliter; limit of detection, 6 IU per milliliter).[15] Additional information about all the trial end points is provided in the Supplementary Methods section and in Table S1.

STATISTICAL ANALYSIS

In primary end-point analysis, the primary comparison was assessed as the difference in the percentage of patients with undetectable HDV RNA at week 24 after the end of treatment between the 10-mg bulevirtide plus peginterferon alfa-2a group and the 10-mg bulevirtide monotherapy group. We calculated that the inclusion of 48 patients in each treatment group would result in a two-sided continuity-corrected 95% confidence interval for the between-group difference in the primary end point that would extend less than 22.5 percentage points from the observed difference. The 2-mg bulevirtide plus peginterferon alfa-2a group was to be of the same size as the two groups that received the 10-mg dose of bulevirtide, and the peginterferon alfa-2a monotherapy group was to include 25 patients, for a total of 175 patients to undergo randomization. The difference in response between the 10-mg bulevirtide plus peginterferon alfa-2a group and the 10-mg bulevirtide monotherapy group, its 95% exact unconditional confidence interval, and P value, which was calculated by means of a two-sided Fisher's exact test, are reported.

For each binary efficacy end point, the percentage of patients with a response and the Clopper–Pearson 95% confidence interval in each group are provided. Missing values were imputed as nonresponse for binary end points and are reported in the Supplementary Appendix. There was no prespecified plan to adjust for multiple comparisons. A P value is reported for the primary end-point analysis; all other comparisons are reported with point estimates and 95% confidence intervals. The widths of the confidence intervals are not adjusted for multiple comparisons and should not be used to infer definitive treatment effects. Prespecified analysis of the primary end point according to subgroups was also conducted. Further details of the statistical analyses are provided in the protocol and the Supplementary Appendix.

RESULTS

PATIENTS

A total of 175 patients underwent randomization, with 25 patients assigned to receive peginterferon alfa-2a monotherapy and 50 patients assigned to each of the other three treatment groups. One patient who had been assigned to the peginterferon alfa-2a group withdrew consent before receiving treatment and was excluded from the analysis (Fig. S2). The demographic and baseline characteristics of the patients were generally balanced across the trial groups and were representative of the population with persons with chronic hepatitis D in regions where this trial was con-

Table 1. Demographic and Clinical Characteristics at Baseline.*

Characteristic	Pegylated Interferon Alfa-2a (N=24)	Bulevirtide, 2 mg + Pegylated Interferon Alfa-2a (N=50)	Bulevirtide, 10 mg + Pegylated Interferon Alfa-2a (N=50)	Bulevirtide, 10 mg (N=50)
Age — yr	41±8	41±9	41±9	40±8
Male sex — no. (%)	18 (75)	33 (66)	35 (70)	38 (76)
Race — no. (%)†				
Asian	4 (17)	3 (6)	4 (8)	4 (8)
Black	0	3 (6)	2 (4)	2 (4)
White	20 (83)	44 (88)	43 (86)	44 (88)
Body-mass index‡				
Mean	26±4	25±4	25±4	26±4
<30 — no. (%)	20 (83)	43 (86)	44 (88)	44 (88)
Cirrhosis — no. (%)§	8 (33)	17 (34)	17 (34)	17 (34)
Liver stiffness — kPa¶	15.8±11.6	12.8±6.4	12.5±7.6	12.7±6.6
HDV RNA — $\log_{10}$ IU/ml	5.2±1.1	5.3±1.4	5.1±1.3	5.5±1.1
HDV genotype — no. (%)				
1	24 (100)	48 (96)	47 (94)	49 (98)
5	0	1 (2)	2 (4)	1 (2)
6	0	1 (2)	0	0
No data	0	0	1 (2)	0
HBV genotype — no. (%)				
A	4 (17)	7 (14)	7 (14)	8 (16)
D	19 (79)	40 (80)	38 (76)	41 (82)
E	0	1 (2)	2 (4)	0
No data or unclassified	1 (4)	2 (4)	3 (6)	1 (2)
HBV DNA — $\log_{10}$ IU/ml‖	1.9±0.8	2.1±1.6	2.0±0.9	2.2±1.5
Positive HBV DNA — no. (%)‖	17 (71)	41 (82)	38 (76)	40 (80)
HBsAg level				
Mean — $\log_{10}$ IU/ml	3.6±0.5	3.7±0.6	3.7±0.7	3.7±0.6
≤1000 IU/ml — no. (%)	3 (12)	5 (10)	7 (14)	5 (10)
≤3000 IU/ml — no. (%)	8 (33)	13 (26)	13 (26)	9 (18)
HBeAg not detected — no. (%)	23 (96)	42 (84)	47 (94)	43 (86)
ALT — U/liter	121±96	108±77	113±99	118±108
Previous interferon therapy — no. (%)	12 (50)	25 (50)	26 (52)	21 (42)
Concomitant NUC therapy — no. (%)**	11 (46)	24 (48)	25 (50)	23 (46)

* Plus–minus values are means ±SD. ALT denotes alanine aminotransferase, HBeAg hepatitis B e antigen, HBsAg hepatitis B surface antigen, HBV hepatitis B virus, HDV hepatitis D virus, and NUC nucleoside or nucleotide analogue.

† Race was determined by the investigators. Race was reported as "other" in one patient (2%) in the 10-mg bulevirtide plus pegylated interferon alfa-2a group.

‡ The body-mass index is the weight in kilograms divided by the square of the height in meters.

§ All the patients with cirrhosis had class A cirrhosis according to the Child–Turcotte–Pugh classification, in which class A (score, 5 or 6) indicates mildly impaired liver function, class B (score, 7 to 9) moderately impaired liver function, and class C (score, 10 to 15) severely impaired liver function. Overall, scores range from 5 to 15, with higher scores indicating worse liver function.

¶ Liver stiffness was assessed by means of transient elastography (FibroScan, Echosens). Levels range from 2.5 to 75.0 kPa (normal range, 2.5 to 7.0), with higher levels indicating increasing severity of liver scarring.

‖ The mean baseline HDV DNA level was calculated in participants who had positive HBV DNA at baseline.

** Concomitant medications were those that were ongoing or that had been stopped on or after the date of the first dose of trial drug and up to the last dose date of the trial drug. In total, 42 to 50% of the patients received tenofovir as a component of their HBV treatment, 8 to 17% received entecavir, and 6% or less received lamivudine.

ducted (Table 1 and Table S2). Most patients were male and White, and the mean age of the patients was 41 years. Approximately one third of the patients had cirrhosis. Nearly all the patients (97%) had HDV genotype 1 infection, and 79% had HBV genotype D infection. A total of 84 patients (48%) had a history of interferon therapy use. Concomitant treatment with nucleoside or nucleotide analogues for chronic hepatitis B was used by 48% of the trial population in similar proportions across the treatment groups.

RESPONSE TO TREATMENT

Undetectable HDV RNA

At the end of treatment, the HDV RNA level was undetectable in 44% of the patients in the 2-mg bulevirtide plus peginterferon alfa-2a group and in 70% of those in the 10-mg bulevirtide plus peginterferon alfa-2a group. By comparison, the HDV RNA level was undetectable at the end of treatment in 21% of the patients in the peginterferon alfa-2a group and in 22% of those in the 10-mg bulevirtide monotherapy group (Fig. 1A).

The primary end point of an undetectable HDV RNA level at week 24 after the end of treatment was met in 4 of 24 patients (17%) in the peginterferon alfa-2a monotherapy group, in 16 of 50 (32%) in the 2-mg bulevirtide plus peginterferon alfa-2a group, in 23 of 50 (46%) in the 10-mg bulevirtide plus peginterferon alfa-2a group, and in 6 of 50 (12%) in the 10-mg bulevirtide monotherapy group (Table 2 and Fig. 1A). The percentage of patients with a primary endpoint response was significantly higher in the 10-mg bulevirtide plus peginterferon alfa-2a group than in the 10-mg bulevirtide monotherapy group (difference, 34 percentage points; 95% confidence interval, 15 to 50; $P<0.001$).

At week 48 after the end of the treatment, 46% of the patients in the 10-mg bulevirtide plus peginterferon alfa-2a group had undetectable HDV RNA, as compared with 12% of those in the 10-mg bulevirtide monotherapy group. The percentage of patients with undetectable HDV RNA appeared to be similar in the 2-mg bulevirtide plus peginterferon alfa-2a group (26%) and the peginterferon alfa-2a group (25%) (Fig. 1A). Among the patients with undetectable HDV RNA at week 24 after the end of treatment, 4 of 4 patients in the peginterferon alfa-2a group, 21 of 23 in the 10-mg bulevirtide plus peginterferon alfa-2a group, 11 of 16 in the 2-mg bulevirtide plus peginterferon alfa-2a group, and 4 of 6 in the 10-mg bulevirtide monotherapy group had an undetectable level at week 48 after the end of treatment. (Data for the two combination-therapy groups are shown in Fig. S5.)

Normalization of Alanine Aminotransferase Level and Composite and Other End Points

At week 24 after the end of treatment, the level of alanine aminotransferase had normalized in 25% of the patients in the peginterferon alfa-2a group, in 42% of those in the 2-mg bulevirtide plus peginterferon alfa-2a group, in 56% of those in the 10-mg bulevirtide plus peginterferon alfa-2a group, and in 30% of those in the 10-mg bulevirtide monotherapy group (Table 2 and Fig. 1B). The composite end point of undetectable HDV RNA and normalization of the alanine aminotransferase level is presented in Table 2 and Figure 1C. Changes in liver stiffness are shown in Figure S3, and additional efficacy end points are presented in Table S3.

HBsAg Loss

HBsAg loss was observed in no patients in the peginterferon alfa-2a group, in 5 patients (10%) in the 2-mg bulevirtide plus peginterferon alfa-2a group, in 2 (4%) in the 10-mg bulevirtide plus peginterferon alfa-2a group, and in 1 (2%) in the 10-mg bulevirtide monotherapy group at 48 weeks after the end of treatment. Most patients who had an undetectable HDV RNA level during the treatment-free follow-up period did not have HBsAg loss (Table 2 and Table S3).

SAFETY

In general, the safety profile of bulevirtide in combination with peginterferon alfa-2a was consistent with the known safety profiles of each drug. The majority of adverse events were of grade 1 or 2 in severity. Vitamin D deficiency, injection-site reaction, headache, dizziness, and pruritus were more commonly reported in the groups receiving bulevirtide than in the peginterferon alfa-2a monotherapy group. Leukopenia, neutropenia, thrombocytopenia, lymphopenia, influenza-like illness, and pyrexia were more commonly reported in the groups receiving peginterferon alfa-2a than in the 10-mg bulevirtide monotherapy group. Combination therapy was linked to a higher incidence of leukopenia, neutropenia, injection-

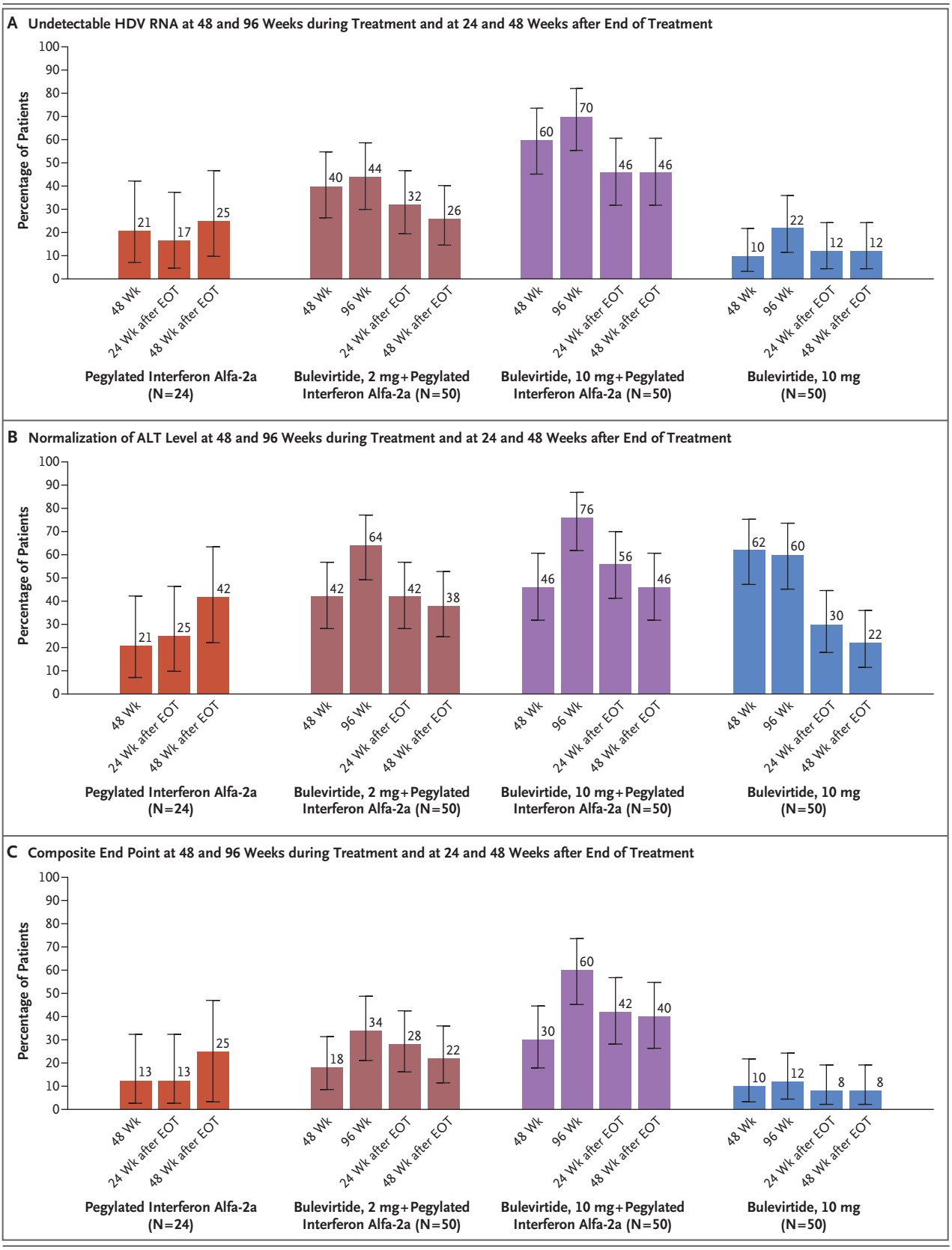

Figure 1 (facing page). Hepatitis D Virus RNA, Normalization of ALT Level, and Composite End Point during Treatment and in the Follow-up Period.

The primary end point of the trial was an undetectable hepatitis D virus (HDV) RNA level at week 24 after the end of treatment (EOT). The end of treatment was at week 48 in the peginterferon alfa-2a group and at week 96 in all the groups that received bulevirtide. Panel A shows the percentage of patients who had an undetectable HDV RNA level; data were analyzed according to the group to which the patients were randomly assigned (planned treatment group). Panel B shows the percentage of patients in whom the alanine aminotransferase (ALT) level had normalized, according to trial group. Panel C shows the percentage of patients who met the composite end-point criteria (undetectable HDV RNA and normalization of the ALT level), according to trial group. In all panels, I bars indicate the 95% confidence interval. No multiplicity adjustment was made for any confidence interval.

site reaction, and anemia than either drug alone. The safety analysis is shown in Table 3.

Serious adverse events during treatment occurred in all four groups, but serious adverse events that were considered by the investigator to be related to trial treatment were attributed to peginterferon alfa-2a. Grade 3 or higher adverse events that were considered by the investigator to be related to bulevirtide were reported in four patients, including injection-site reaction and decreased neutrophil count (in one patient each in the 2-mg bulevirtide plus peginterferon alfa-2a group) and increased alanine aminotransferase level and increased aspartate aminotransferase level (in one patient each in the 10-mg bulevirtide plus peginterferon alfa-2a group). In three patients, bulevirtide treatment was discontinued early owing to an adverse event. One patient in the 10-mg bulevirtide monotherapy group had myalgia that was deemed by the investigator to be related to treatment, one patient in the 10-mg bulevirtide plus peginterferon alfa-2a group had drug-induced liver injury that was deemed by the investigator to be related to peginterferon alfa-2a, and one fatal case of anaplastic astrocytoma occurred in the 2-mg bulevirtide plus peginterferon alfa-2a group, which the investigator considered to be unrelated to treatment.

Dose-dependent elevations in bile acids were observed as expected in the groups receiving bulevirtide (Fig. S4). No apparent relationship was observed between bile acid levels and any symptoms, including pruritus (Fig. S8).

SAFETY DURING POST-TREATMENT PERIOD

Post-treatment hepatic events are described in Table S5. Elevations in alanine aminotransferase and aspartate aminotransferase levels were reported in 10 to 16% of the patients in the bulevirtide plus peginterferon alfa-2a groups; in the 10-mg bulevirtide monotherapy group, an elevation in the alanine aminotransferase was observed in 28% of the patients, and an elevation in the aspartate aminotransferase level was observed in 22%. Most of the events were asymptomatic, were associated with HDV RNA rebounds, and resolved without treatment. An increase in the bilirubin level occurred in 6% of the patients in the 10-mg bulevirtide plus peginterferon alfa-2a group and in 10% of those in the 10-mg bulevirtide monotherapy group, including in two patients in the monotherapy group who had jaundice.

Varices bleeding was suspected in one patient in the 10-mg bulevirtide plus peginterferon alfa-2a group. One death from esophageal varices hemorrhage occurred in a patient with hepatocellular carcinoma in the 10-mg bulevirtide plus peginterferon alfa-2a group. The effect of bulevirtide antidrug antibodies on efficacy and safety is discussed in the Supplementary Appendix.

DISCUSSION

In this phase 2b trial, treatment with bulevirtide daily for 96 weeks, combined with weekly peginterferon alfa-2a therapy for the first 48 weeks, resulted in a higher percentage of patients meeting the primary end point of undetectable HDV RNA at 24 weeks after the end of treatment than bulevirtide alone. Among the regimens that we evaluated, the highest percentages of patients with an undetectable HDV RNA level at the end of treatment and at weeks 24 and 48 after the end of treatment were observed in the 10-mg bulevirtide plus peginterferon alfa-2a group. Our results indicate that a regimen of finite duration for chronic hepatitis D led to a sustained undetectable HDV RNA response, as measured by a highly sensitive HDV RNA assay, beyond 24 weeks after the end of treatment.[16] This response appeared to be maintained from 24 to 48 weeks after the end of treatment — a finding that supports the concept that sustained undetectable HDV RNA for at least 1 year after treatment is possible in patients with chronic hepatitis D who have been treated with a finite duration of therapy

Table 2. Efficacy End Points.*

Response	Pegylated Interferon Alfa-2a (N = 24)	Bulevirtide, 2 mg + Pegylated Interferon Alfa-2a (N = 50)	Bulevirtide, 10 mg + Pegylated Interferon Alfa-2a (N = 50)	Bulevirtide, 10 mg (N = 50)
Undetectable HDV RNA				
At wk 48				
No. of patients	5	20	30	5
Percentage of patients (95% CI)	21 (7–42)	40 (26–55)	60 (45–74)	10 (3–22)
At wk 96				
No. of patients	NA	22	35	11
Percentage of patients (95% CI)	—	44 (30–59)	70 (55–82)	22 (12–36)
At wk 24 after EOT				
No. of patients	4	16	23	6
Percentage of patients (95% CI)	17 (5–37)	32 (20–47)	46 (32–61)	12 (5–24)
At wk 48 after EOT				
No. of patients	6	13	23	6
Percentage of patients (95% CI)	25 (10–47)	26 (15–40)	46 (32–61)	12 (5–24)
Normalization of ALT level				
At EOT				
No. of patients	5	32	38	30
Percentage of patients (95% CI)	21 (7–42)	64 (49–77)	76 (62–87)	60 (45–74)
At wk 24 after EOT				
No. of patients	6	21	28	15
Percentage of patients (95% CI)	25 (10–47)	42 (28–57)	56 (41–70)	30 (18–45)
At wk 48 after EOT				
No. of patients	10	19	23	11
Percentage of patients (95% CI)	42 (22–63)	38 (25–53)	46 (32–61)	22 (12–36)
HBsAg loss				
At wk 24 after EOT — no. (%)	0	4 (8)	2 (4)	0
At wk 48 after EOT — no. (%)	0	5 (10)	2 (4)	1 (2)

* No multiplicity adjustment was made for any confidence interval. The end of treatment (EOT) was at week 48 in the peginterferon alfa-2a group and at week 96 in all the groups that received bulevirtide. CI denotes confidence interval, and NA not applicable.

of at least 96 weeks, including 48 weeks of peginterferon alfa-2a therapy.

Most patients with an undetectable HDV RNA level during the treatment-free follow-up period did not have HBsAg loss, which suggests that an undetectable HDV RNA level can be achieved and sustained without HBsAg loss.[17] These results support the possibility that HDV infection can resolve without functional clearance of HBV infection.[17] Observation of this treatment effect lends support to the possibility of a synergistic effect when HDV entry inhibition with bulevirtide is combined with control of cell division–mediated HDV spread with peginterferon alfa-2a.[18,19]

In this trial, 70% of the patients in the 10-mg bulevirtide plus peginterferon alfa-2a group had an undetectable HDV RNA level at the end of treatment, which differs from results of the Hep-Net International Delta Hepatitis Interventional Trial II (HIDIT-II), in which 33 to 48% of the patients had undetectable levels after 96 weeks of peginterferon alfa-2a therapy, with or without tenofovir disoproxil, and from the results of the phase 3 MYR 301 trial, which

Table 3. Safety Outcomes.*

Event	Pegylated Interferon Alfa-2a (N=24)	Bulevirtide, 2 mg + Pegylated Interferon Alfa-2a (N=50)	Bulevirtide, 10 mg + Pegylated Interferon Alfa-2a (N=50)	Bulevirtide, 10 mg (N=50)
	number of patients (percent)			
Any adverse event				
Overall	22 (92)	49 (98)	50 (100)	42 (84)
Grade ≥3 event	13 (54)	27 (54)	30 (60)	10 (20)
Bulevirtide-related adverse event				
Overall	NA	25 (50)	21 (42)	22 (44)
Grade ≥3 event	NA	2 (4)	2 (4)	0
Pegylated interferon alfa-2a–related adverse event				
Overall	21 (88)	49 (98)	50 (100)	NA
Grade ≥3	13 (54)	26 (52)	26 (52)	NA
Serious adverse event				
Overall	3 (12)	3 (6)	8 (16)	2 (4)
Related to bulevirtide	NA	0	0	0
Related to pegylated interferon alfa-2a	1 (4)	2 (4)	1 (2)	NA
Adverse event leading to premature discontinuation				
Related to bulevirtide	NA	1 (2)	1 (2)	1 (2)
Related to pegylated interferon alfa-2a	1 (4)	3 (6)	2 (4)	NA
Death	0	1 (2)	1 (2)†	0
Adverse events in ≥10% of patients in any group‡				
Leukopenia	15 (62)	36 (72)	35 (70)	9 (18)
Neutropenia	15 (62)	34 (68)	40 (80)	7 (14)
Thrombocytopenia	16 (67)	32 (64)	31 (62)	9 (18)
Influenza-like illness	10 (42)	22 (44)	20 (40)	2 (4)
Lymphopenia	7 (29)	12 (24)	15 (30)	5 (10)
Vitamin D deficiency	3 (12)	9 (18)	13 (26)	13 (26)
ALT increased	8 (33)	5 (10)	12 (24)	5 (10)
AST increased	8 (33)	6 (12)	10 (20)	3 (6)
GGT increased	6 (25)	2 (4)	7 (14)	3 (6)
Injection-site reaction	0	11 (22)	11 (22)	5 (10)
Pyrexia	6 (25)	8 (16)	13 (26)	0
Headache	2 (8)	7 (14)	7 (14)	7 (14)
Asthenia	3 (12)	7 (14)	6 (12)	4 (8)
Activated partial thromboplastin time prolonged	3 (12)	6 (12)	5 (10)	1 (2)
Anemia	1 (4)	4 (8)	6 (12)	1 (2)
Weight decreased	3 (12)	3 (6)	4 (8)	2 (4)
Covid-19	0	3 (6)	6 (12)	3 (6)
Dizziness	1 (4)	3 (6)	2 (4)	5 (10)
Hyperbilirubinemia	3 (12)	0	2 (4)	4 (8)
Pruritus	0	2 (4)	5 (10)	3 (6)

* The adverse events that are reported here include any event that was reported on or after the date of the start of the trial drug up to 30 days after permanent discontinuation of the trial drug or that led to premature discontinuation of the trial drug. Relatedness to bulevirtide or pegylated interferon alfa-2a was determined by the investigator. AST denotes aspartate aminotransferase, Covid-19 coronavirus disease 2019, and GGT γ-glutamyltransferase.

† Death occurred in the post-treatment period.

‡ The *Medical Dictionary for Regulatory Activities*, version 26.0, was used to code these adverse events. Leukopenia included the term white-cell count decreased, neutropenia included the term neutrophil count decreased, and thrombocytopenia included the term platelet count decreased. A full description of injection-site reaction terms is provided in the Supplementary Appendix.

showed that HDV RNA was undetectable in 20 to 36% of the patients after 96 weeks of bulevirtide monotherapy.[10,20] An increase in the percentage of patients with an undetectable HDV RNA level was observed in both combination-therapy groups after peginterferon alfa-2a therapy was discontinued at week 48 and patients were continuing to receive bulevirtide alone. This finding highlights the potential for bulevirtide monotherapy to maintain response after the use of combination therapy.

Leukopenia, neutropenia, injection-site reaction, and anemia occurred more often with combination therapy than with either treatment alone. The nature and severity of the adverse events that were attributed to peginterferon alfa-2a or bulevirtide in patients who received combination therapy were similar to those of each therapy individually. No serious adverse event that was considered by the investigator to be related to bulevirtide occurred during the treatment period, and discontinuation of bulevirtide was rare. Hepatitis exacerbation is a known risk after the cessation of bulevirtide treatment and was closely monitored in this trial.[21] In the post-treatment period, increases in the alanine aminotransferase level were observed, mostly in association with HDV rebound; however, most of these events were asymptomatic and resolved spontaneously.

Our trial has several limitations. Foremost, this trial was not designed to compare the two doses of bulevirtide and therefore lacked the sample size to allow for formal comparisons. In addition, although the trial included a peginterferon alfa-2a monotherapy group, it was not powered to allow for comparison. The response that we observed with peginterferon alfa-2a monotherapy was similar to historical data.[11,22,23] Monotherapy with 2 mg of bulevirtide was not evaluated. There was a lack of blinding to the treatment intervention due to ethical reasons, such as daily placebo injections; nevertheless, since efficacy end points were derived from objective laboratory measurements, the integrity of the trial data was maintained.

The trial population consisted almost entirely of patients with HDV genotype 1 or HBV genotype A or D, which may limit the generalizability of the results. However, HDV genotype 1 represents nearly 90% of the cases of chronic hepatitis D worldwide, and HBV genotypes A and D are also found worldwide.[1] On the basis of in vitro data that were obtained with all recombinant genotype combinations of HBV genotypes A through H and HDV genotypes 1 through 8 and the lack of viral resistance reported thus far, bulevirtide is expected to possess potent pangenotypic activity.[24,25] In addition, a conservative approach of imputing missing data as treatment failure was taken for the efficacy end points. Finally, the post-treatment follow-up period was limited to 48 weeks. Although this duration was longer than the follow-up in most other interventional trials in this disease, it did not allow for the assessment of longer-term clinical liver-related outcomes or late relapses.

In this phase 2b trial involving patients with chronic hepatitis D, combination therapy with 10 mg of bulevirtide daily for 96 weeks, plus peginterferon alfa-2a for the first 48 weeks, resulted in a significantly higher percentage of patients with an undetectable HDV RNA level at 24 weeks after the end of treatment than monotherapy with 10 mg of bulevirtide for 96 weeks.

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APPENDIX

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