FINAL ABBREVIATED CLINICAL STUDY REPORT

Study Title:	Pharmacoepidemiology study to define the long-term safety profile of tenofovir disoproxil fumarate (TDF, Viread) and describe the management of TDF-associated renal and bone toxicity in Chronic Hepatitis B (CHB)-infected adolescents aged 12 to <18 years in Europe
Name of Test Drug:	Tenofovir Disoproxil Fumarate
Dose and Formulation:	Viread 300-mg tablet
Indication:	Chronic Hepatitis B infection
Sponsor:	Gilead Sciences Ireland UC, Carrigtohill, County Cork T45 DP77, Ireland
Study No.:	GS-EU-174-1403
Phase of Development:	Phase 4
IND No.: **EudraCT No.:**	Not Applicable 2014-004939-39
ClinicalTrials.gov Identifier	NCT02479880
Study Start Date:	03 July 2015 (First Subject Screened)
Study End Date:	11 April 2018 (Last Subject Last Observation for the Primary Endpoint)
Principal or Coordinating Investigator:	Name: Tara MacCannell, PhD MSc Affiliation: Gilead Sciences, Inc.
Gilead Responsible Medical Monitor:	Name: John Flaherty, PharmD Telephone: PPD Fax: PPD
Report Date:	25 September 2018
Previous Report Date(s):	23 January 2017 (18 month study enrollment report)

CONFIDENTIAL AND PROPRIETARY INFORMATION

This study was conducted in accordance with the guidelines of Good Clinical Practice, including archiving of essential documents.

STUDY SYNOPSIS

Study GS-EU-174-1403
Gilead Sciences Ireland UC
Carrigtohill, County Cork
T45 DP77, Ireland

Title of Study: Pharmacoepidemiology study to define the long-term safety profile of tenofovir disoproxil fumarate (tenofovir DF, Viread) and describe the management of Viread-associated renal and bone toxicity in Chronic Hepatitis B (CHB)-infected adolescents aged 12 to <18 years in Europe
Investigators: Multicenter Study
Study Centers: 1 site each in Belgium, France, Greece, Italy, and the United Kingdom, and 3 sites each in Spain and Romania
Publications: There were no publications at the time of this CSR
Study Period: 03 July 2015 (First Subject Screened) 11 April 2018 (Last Subject Last Observation for the Primary Endpoint)
Phase of Development: Phase 4

Objectives:

The primary objective of this study was as follows:

To characterize the long term (i.e., 96 weeks of follow up) bone safety profile of open-label Viread treatment in adolescents with chronic hepatitis B (CHB). This includes prospectively evaluating and comparing the bone mineral density (BMD) change between adolescents with CHB 12 to < 18 years of age who were treated with Viread in European treatment centers who were assigned to one of two schedules for renal and bone laboratory monitoring and BMD measurement. Primary study outcome was the percent changes in BMD from Baseline through study Week 96.

The secondary objectives of this study were as follows:

- To describe the demographics and disease characteristics of adolescents with CHB treated with Viread
- To document all serious adverse drug reactions (SADR) and all renal- and bone-related adverse events (AEs), including renal and bone laboratory abnormalities
- To determine the time to diagnosis of renal and bone AEs and document the resulting patient management and outcome(s)
- To assess the clinical management and outcomes of renal- and bone-related ≥ Grade 3 laboratory markers and clinical serious adverse events (SAEs)
- To assess the efficacy and tolerability of Viread in adolescents with CHB
- To assess the use of oral vitamin D, calcium, and phosphate supplementation and explore the association between supplement use and rates of bone and renal AEs
- To describe reasons for discontinuation of Viread.

Methodology:

This was an interventional study involving two assigned monitoring groups of adolescents with CHB who received treatment with open-label, market-authorized Viread and were followed prospectively to a primary endpoint at Week 96. Subjects were assigned to one of two monitoring groups (Group 1 or 2); subjects in both groups followed preset schedules for renal, bone, and bone biomarker monitoring. Group 1 received more frequent BMD, bone biomarker, and renal function monitoring compared to Group 2 subjects who were followed according to the monitoring recommendations contained in the current Summary of Product Characteristics (SmPC) over the 96 week study period. Subjects were randomly assigned but not blinded to one of the two monitoring groups.

Number of Subjects (Planned and Analyzed):
Planned: 100 subjects to receive treatment with Viread and assigned to one of two monitoring groups (50 subjects in each group)
Analyzed: 30 subjects (15 subjects in each group) were analyzed as a result of the Pharmacovigilance Risk Assessment Committee's (PRAC) request to cancel the study due to low enrollment

Diagnosis and Main Criteria for Inclusion: At screening, subjects aged 12 to <16 years of age at the time of enrollment with chronic HBV (e.g., HBsAg-positive for at least 6 months), and weighing ≥35kg were eligible for the study. Subjects must have been naïve to Viread, but could have previously received interferon and/or any oral anti-HBV nucleoside/nucleotide therapy. Subjects experienced on interferon must have discontinued therapy for a minimum of 6 months; treatment-experienced subjects receiving oral anti-HBV nucleoside/nucleotide treatment at screening must continue their current treatment regimen until Viread was initiated; subjects previously treated with an oral anti-HBV nucleoside/nucleotide were to initiate Viread without a treatment washout period to prevent ALT flare. Pregnant or breastfeeding females were not eligible for study participation.

Duration of Treatment: 96 weeks

Test Product, Dose, Mode of Administration: 300 mg oral tenofovir disoproxil fumarate tablets (Viread), which is equivalent to 245 mg tenofovir disoproxil, following enhanced patient monitoring protocol for bone and renal biomarkers

Reference Therapy, Dose, Mode of Administration: The reference therapy was 300mg Viread following local standards of care for patient monitoring for renal laboratory parameters

Criteria for Evaluation:

Safety: The primary safety endpoint was the indication of a cumulative incidence of a ≥4% decrease from Baseline in bone mineral density (BMD) of the spine or whole body through Week 96 performed by dual energy X-ray absorptiometry (DXA). Secondary endpoints included bone and renal AEs. Bone AEs may include reports of bone pain or fractures. Renal AEs may include reductions in creatinine clearance, and clinical and laboratory evidence of Grade 3 or greater renal tubulopathy or toxicity (e.g. proximal renal tubulopathy, Fanconi syndrome). Bone and renal AEs, concomitant medications (including dietary supplements) and specified clinical laboratory tests were collected each time subjects from Group 1 or Group 2 returned for scheduled monitoring by the study site over a 96-week period of evaluation.

Efficacy: An efficacy analysis was to be conducted after the last assigned subject reached Week 96. The analysis was to have evaluated the difference in the proportion of all subjects (Groups 1 and 2 data were pooled, as all subjects were exposed to the same study drug) achieving HBV DNA < 400 copies/mL (69 IU/mL) and a normal level of serum ALT at Week 96.

Pharmacokinetics: Not Applicable

Statistical Methods:

Safety: The primary safety analysis was to be performed after the last subject reached Week 96 on Viread treatment among both monitoring groups. The primary analysis evaluated quantitative percent changes in spine or whole body BMD compared to baseline assessments through serial measurements of DXA scanning. A key secondary study objective was to assess within-group and between-group differences in renal laboratory parameter changes (e.g., creatinine clearance using the Schwartz formula, glucosuria, proteinuria) suggestive of renal toxicity. To evaluate the time to detection of renal or bone AEs among the assigned monitoring groups, estimations of incidence rates (events over person time on Viread) and time to event analysis were proposed.

Efficacy: The evaluation of other clinically relevant endpoints such as virologic response, serologic status, and development of Viread resistance were to also be assessed as secondary study outcomes for each monitoring group and as a combined cohort.

Refer to GS-EU-174-1403 Statistical Analysis Plan for additional details on the analysis to evaluate between group differences.

SUMMARY OF RESULTS:

Subject Disposition and Demographics:

A total of 30 of the planned 100 subjects were randomized to this study (15 subjects in Monitoring Group 1, and 15 subjects in Monitoring Group 2). At the request of PRAC, the study was permanently discontinued early due to low subject enrollment. There were 11 study enrollment sites among 7 EU member states with 1 site each in Belgium, France, Greece, Italy, and the UK, and 3 sites each in Romania and Spain.

All subjects received at least one dose of study drug (Viread). Demographic and baseline characteristics were similar between monitoring groups. Nineteen (63.3%) subjects (11 subjects from Monitoring Group 1, 8 subjects from Monitoring Group 2) were randomized when subjects were between the ages of 12 and 14 years. The remaining 11 subjects (36.7%) were either 15 or 16 years of age at the time of randomization.

The composition of the study included 20 males (66.7%) and 10 females (33.3%). The study population included 11 subjects (36.7%) who were white, 10 black subjects (33.3%), and 7 Asian subjects (23.3%). All subjects were confirmed to be HBsAg positive at baseline. The majority of subjects were infected with HBV genotype D (60.0%, or 18 subjects), followed by genotypes A and E, with 10% (3 subjects) in each of these groups. Overall, 67.7% of subjects were treatment naïve (13 in Monitoring Group 1 and 7 in Monitoring Group 2). The mean baseline HBV DNA was $\log_{10}$7.85 copies/mL, mean baseline ALT was 56 U/L, and 70.0% of subjects (21 of 30 subjects) were HBeAg+ at baseline, with no statistical differences between monitoring groups.

Efficacy Results:

The primary efficacy endpoint was the proportion with HBV DNA <400 copies/mL (<69 IU/mL) at Week 96; however, only 3 subjects could be assessed for this endpoint. Of the 30 enrolled study subjects, 24 subjects completed 48 weeks of treatment and were assessed for efficacy at this time point, while only 4 subjects (2 subjects from each monitoring group) completed the 96 week study period (note that only 3 subjects who completed the study had HBV DNA data available at Week 96). A fifth subject provided Week 96 efficacy and safety data but was routed to early study discontinuation procedures, as determined by the site. At Week 48, 12 of 24 subjects (50.0%) had HBV DNA values < 400 copies/mL. The proportion of subjects achieving viral suppression remained constant after 48 weeks among subjects with available data.
One subject from Monitoring Group 1 completed the study through Week 96 and achieved the HBV DNA endpoint of ≤400 copies/mL. The second subject from Monitoring Group 1 who completed the study provided a HBV DNA sample which was unable to be analyzed. In Monitoring Group 2, of the 2 subjects with available viral load data at Week 96, 1 subject achieved undetectable HBV DNA levels, and the other subject had HBV DNA >400 copies/mL. With few data to report, the efficacy endpoint for the study could not be described for 86.8% (n=26) subjects.

Sixteen subjects presented with normal ALT (i.e., at or below the upper limit of normal [≤ULN] and 14 subjects presented with ALT >ULN at baseline. Among those initiating the study with normal ALT levels, 13 subjects also had baseline HBV DNA <400 copies/mL and all of these subjects maintained normal ALT levels at Week 48. By Week 48, 63.6% of subjects (n=11) demonstrated HBV DNA <400 copies/mL among those initiating the study with abnormal ALT levels. The data were too limited at Week 96 study to adequately describe changes in viral load at this primary efficacy time point.

At Week 96, compared with baseline, the median HBV DNA (IQR Q1, Q3) $\log_{10}$ decrease was -5.57 (-6.70, -3.47) copies/mL and represented by only 4 subjects, hence subject data were too few to interpret. The available data indicate a steady decline from a median (IQR Q1, Q3) HBV DNA of $\log_{10}$ 8.92 (6.17, 9.63) copies/mL to $\log_{10}$ 2.66 (2.06, 3.57) copies/mL from baseline to Week 48 based on 30 and 24 total subjects, respectively. The median Week 96 HBV DNA results pooled across all groups (n=4) was $\log_{10}$2.38 (2.06, 3.88) copies/mL, and despite few subjects, potentially points to further decreases in HBV viral load.

Among the 21 subjects who were HBeAg positive at baseline, HBeAg loss and seroconversion were achieved in 1 subject from Monitoring Group 1 at Week 48, and in 2 subjects at Weeks 72 and 96 in Monitoring Group 2, respectively.

Median (IQR Q1, Q3) adherence to Viread was 93.8% (87.9, 97.7) during the study. There were 20 subjects (66.7%) across both monitoring groups who demonstrated ≥90% adherence to Viread based on pill counts. Treatment adherence less than 80% occurred in 3 subjects (10.0%). All four subjects completing the study demonstrated >80% treatment adherence.

Pharmacokinetics/Pharmacodynamics Results: Not applicable

Safety Results:

No subjects discontinued Viread treatment as a result of an AE. There were no subjects in Monitoring Group 1 (n=15) who experienced treatment emergent bone-related AEs compared with 3 of 15 subjects (20.0%) reporting bone-related AEs in Monitoring Group 2. There were no treatment emergent renal AEs reported in either monitoring group, and therefore no reports of clinical management of renal-related AEs or outcomes.

Treatment emergent laboratory abnormalities were few, and overall no Grade 4 abnormalities were detected. No subjects across the monitoring groups experienced either on-treatment or post-treatment ALT flare. No subject died during the study.

Treatment Emergent Bone-Related Adverse Events

Three treatment emergent bone-related adverse events were reported in 3 subjects. One subject experienced a right forearm fracture on Day 327 which was judged by the investigator as being unrelated to Viread. Two subjects experienced decreased bone mineral density, AEs which were considered related to Viread exposure by the investigator. One subject had a Grade 1 AE (Study Day 335) and the other subject had a Grade 2 AE (Study Day 642) with regards to severity of BMD decrease; the detection of these AEs coincided with receiving DXA scans. For these 2 subjects, the resolution of these AEs was missing or unknown by the end of study. There was no reported concomitant use of oral supplements – vitamin D, calcium, or phosphorus based formulations, during the study to mitigate bone density loss in the subject with a Grade 1 bone event. Bone mineral loss observed in the subject with a Grade 2 event had been supplemented with oral vitamin D and calcium during the study but these had been discontinued several months prior to the AE. Across all study subjects, 6 were prescribed oral supplements, 3 in each monitoring group. Five of these 6 subjects were given supplements to augment vitamin D or calcium levels.

Treatment Emergent Grade 3 or 4 Adverse Events

There were 4 treatment emergent Grade 3 AEs in 4 subjects and no Grade 4 AEs. Three of these AEs were reported in 3 subjects experiencing low vitamin D levels and were classified as unrelated to Viread treatment. The dose of Viread was not changed nor treatment suspended. The fourth Grade 3 AE was a spontaneous abortion at 8 weeks gestation in a PPD female subject, which was also reported as a serious adverse event after the subject fell in her home. Viread treatment had been discontinued 2 weeks prior to this adverse event when the pregnancy was confirmed. There were no further investigations or updates for this case at the time of study close out.

Treatment Emergent Serious Adverse Events

There were a total of 3 treatment emergent serious adverse events occurring in 3 subjects across all monitoring groups and none of the SAEs were considered related to study drug treatment by the investigator. In Monitoring Group 1, a PPD male subject was diagnosed with a right ankle anterior astragalus calcification (calcinosis) via MRI (Study Day 432). The subject was hospitalized to repair the defect and discharged, and followed with post-operative physiotherapy.

In Monitoring Group 2, a PPD female subject experienced intermittent episodes of malaise leading to multiple hospitalizations both prior to and during the study. Etiology was unknown and classified as a pre-existing condition. A third treatment emergent SAE occurred in a PPD female subject, also from Monitoring Group 2, who experienced a spontaneous abortion after a fall in her home (Study Day 197). Viread treatment had been discontinued at the time the pregnancy was confirmed 2 weeks prior to the miscarriage.

Percent Changes in Bone Mineral Density from Baseline to Week 96

No subject met or exceeded the pre-specified endpoint of a 4% decrease in spine or whole body BMD at Week 96. As the study was canceled, only 7 of the 30 subjects (23.3%) had DXA scans performed within the 96 week study window. There were 2 subjects with a ≥4% decrease in either spine or whole body BMD measurements prior to Week 96– each of these subject's BMD assessments improved to less than a 4% decrease with subsequent DXA scans. In Monitoring Group 1, where subjects were scheduled for BMD assessments every 6 months after baseline, 1 subject was detected with a ≥4% decrease in whole body BMD at 48 weeks, and another subject showed a single signal of a ≥4% decrease in spine BMD at 24 weeks. In Monitoring Group 2, where subjects were scheduled for BMD assessment annually after baseline, no changes ≥4% was detected at any time point. From this sample, 2 subjects had a cumulative incidence of at least a 4% decrease in spine or whole body BMD from baseline, but these signals did not persist to Week 96 from available data. Given these results, tt is not feasible to draw clear interpretations with these sparse data.

For spine BMD, 14 of 15 subjects (93.3%) from Monitoring Group 1 and 10 of 15 subjects (66.7%) from Monitoring Group 2 had normal z-scores at baseline (i.e. z-score > -1). Overall, there were 6 subjects (20.0%) with low BMD at spine at baseline which was defined for this study as having a BMD z-score between -1 and -2. Five of these 6 subjects were in Monitoring Group 2. Mean spine BMD z-scores were not statistically significant (p=0.17) between groups at baseline. At Week 48, 24 subjects had spinal DXA results available and 60.0% were reported to have z-scores greater than -1. Among the 7 subjects completing DXA scans within the 96 week assessment window, all reported z-scores were greater than -1. Similar trends were observed with available data for whole body BMD z-scores at baseline, Week 48 and Week 96. No statistically significant differences arose between monitoring groups. Two subjects reported z-scores less than -2, 1 from each monitoring group, which were captured up until Week 72 at early study discontinuation. The available data are too few to draw conclusions.

Pregnancy

In Monitoring Group 2, a PPD female was confirmed by ultrasound to be 5 weeks pregnant (Study Day 184). Viread treatment was discontinued at the time the pregnancy was confirmed. The pregnancy terminated prematurely – the subject experienced a spontaneous abortion (Grade 3 SAE) after a fall in her home approximately 2 weeks after the pregnancy was confirmed.

Other Outcomes

As the study was prematurely discontinued before the majority of subjects could complete the target of 96 weeks of monitoring, the study objective to measure the benefit of enhanced subject monitoring vs standard of care could not be adequately assessed due to a lack of data.

CONCLUSIONS:

The conclusions from the final analysis of Study GS-EU-174-1403 following premature discontinuation of the study are as follows:

- Treatment with Viread was generally safe and well-tolerated with no subjects discontinuing treatment for an adverse event
- No renal adverse events were detected
- No Grade 4 adverse events were reported
- Few bone-related treatment emergent adverse events were reported which were mild to moderate in severity
- Few subjects had BMD results available at Week 96 to draw conclusions, and those with ≥4% decreases showed subsequent resolution with repeated DXA scanning.
- Four subjects met the criteria for study completion and 3 had reportable HBV DNA results at Week 96. Two of the 3 subjects had HBV DNA ≤400 copies/mL and 1 subject had HBV DNA >400 copies/mL. Too few subjects met the study endpoint to assess efficacy.

Based on the limited data available from this analysis, the primary study objective to evaluate the effect of using enhanced or standard of care monitoring strategies to prevent or provide early detection of bone or renal AEs could not be adequately addressed.