# Efficacy and safety of extended duration letermovir prophylaxis in recipients of haematopoietic stem-cell transplantation at risk of cytomegalovirus infection: a multicentre, randomised, double-blind, placebo-controlled, phase 3 trial

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## Summary

**Background** In a pivotal phase 3 trial of cytomegalovirus prophylaxis with letermovir for up to 100 days after allogeneic haematopoietic stem-cell transplantation (HSCT), 12% of participants developed clinically significant cytomegalovirus infection after letermovir was discontinued. We aimed to evaluate the efficacy and safety of extending the duration of letermovir prophylaxis for clinically significant cytomegalovirus infection from 100 days to 200 days following HSCT.

**Methods** We conducted a multicentre, randomised, double-blind, placebo-controlled, phase 3 trial at 32 sites in six countries (France, Germany, Italy, Japan, the UK, and the USA). Cytomegalovirus-seropositive HSCT recipients (aged ≥18 years) who had received letermovir prophylaxis for up to 100 days following HSCT and who remained at high risk of late clinically significant cytomegalovirus infection (with no previous history of clinically significant cytomegalovirus infection, defined as initiation of pre-emptive therapy for documented cytomegalovirus viraemia, onset of cytomegalovirus end-organ disease, or both) were eligible. Participants were randomly assigned (2:1) to receive either an additional 100 days (ie, a total of 200 days; letermovir group) of oral or intravenous letermovir 480 mg once daily, adjusted to 240 mg once daily for participants on cyclosporin A, or 100 days of a placebo comparator for letermovir (ie, a total of 100 days of letermovir; placebo group), following HSCT. Randomisation was done using a central interactive response technology system, stratified by study centre and haploidentical donor (yes or no). Participants, investigators, and sponsor personnel were masked to the treatment allocation. The primary efficacy endpoint was the proportion of participants from randomisation to week 28 (200 days after HSCT) with clinically significant cytomegalovirus infection, analysed using the full analysis set population (ie, those who received at least one dose of study intervention). Safety was analysed in all participants as treated (ie, those who received at least one dose according to the study intervention they were assigned to). This study is registered with ClinicalTrials.gov, NCT03930615, and is complete.

**Findings** Between June 21, 2019, and March 16, 2022, 255 patients were screened for eligibility and 220 (86%) were randomly assigned (145 [66%] in the letermovir group and 75 [34%] in the placebo group). Between randomisation and week 28, four (3%) of 144 participants in the letermovir group and 14 (19%) of 74 in the placebo group developed clinically significant cytomegalovirus infection (treatment difference −16·1% [95% CI −25·8 to −6·5]; p=0·0005). The most common adverse events among participants in the letermovir group versus the placebo group were graft-versus-host disease (43 [30%] *vs* 23 [31%]), diarrhoea (17 [12%] *vs* nine [12%]), nausea (16 [11%] *vs* 13 [18%]), pyrexia (13 [9%] *vs* nine [12%]), and decreased appetite (six [4%] *vs* nine [12%]). The most frequently reported serious adverse events were recurrent acute myeloid leukaemia (six [4%] *vs* none) and pneumonia (three [2%] *vs* two [3%]). No deaths were considered to be drug-related by the investigator.

**Interpretation** Extending the duration of letermovir prophylaxis to 200 days following HSCT is efficacious and safe in reducing the incidence of late clinically significant cytomegalovirus infection in patients at risk.

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## Introduction

Cytomegalovirus is a common viral infection in recipients of allogeneic haematopoietic stem-cell transplantation (HSCT) and is associated with substantial morbidity and mortality.[1–3] The period of highest risk for cytomegalovirus reactivation, leading to clinically significant cytomegalovirus infection (ie, cytomegalovirus infection requiring pre-emptive

## Research in context

### Evidence before this study

The results of a large, global, phase 3 study (the P001 study) showed that letermovir was well tolerated and superior to placebo in reducing the incidence of clinically significant cytomegalovirus infection until week 24 (around 200 days) following haematopoietic cell transplantation (HSCT). These results supported the approval of letermovir for cytomegalovirus prophylaxis in cytomegalovirus-seropositive adult recipients of allogeneic HSCT until day 100 following HSCT. However, in the P001 study, an increased incidence of clinically significant cytomegalovirus infection (12·1%) was observed between 100 days and 200 days after letermovir was discontinued at 100 days. We searched PubMed, without language restrictions, for clinical trials evaluating an extended duration of cytomegalovirus prophylaxis with letermovir, published from the time of approval of letermovir (Nov 1, 2017) to the start date of this trial (June 21, 2019), using the following search terms: "letermovir" AND "extended duration" AND "clinical trial". Although we did not identify any clinical trials, multiple single-centre retrospective studies have reported the benefit of extending letermovir beyond 100 days following HSCT in select populations of HSCT recipients who remain at high risk of clinically significant cytomegalovirus infection. A 2022 systematic literature review and meta-analysis of 48 unique observational studies, most of which were single-centre, concluded that primary prophylaxis with letermovir (ranging in duration between 79 days and 191 days) was efficacious in reducing cytomegalovirus-related complications and overall mortality in HSCT recipients beyond 200 days. However, to our knowledge, there were no prospective, randomised, placebo-controlled trials to establish this benefit.

### Added value of this study

Although the evidence base for the safety and efficacy of primary prophylaxis with letermovir administered for up to 100 days following HSCT is well established, questions remain regarding the benefits of extending the duration of prophylaxis in the subpopulation of HSCT recipients at risk of clinically significant cytomegalovirus infection beyond 100 days. The results of this multicentre, randomised, double-blind, placebo-controlled, phase 3 trial support extending the duration of letermovir prophylaxis for up to 200 days in patients who remain at risk of late clinically significant cytomegalovirus infection; letermovir was generally efficacious and safe when administered for 200 days following HSCT.

### Implications of all the available evidence

Cytomegalovirus prophylaxis with letermovir is efficacious and safe in cytomegalovirus-seropositive adult recipients of HSCT for up to 100 days following HSCT. Extending the duration of letermovir prophylaxis to 200 days is a patient-centric, risk-adapted approach to letermovir prophylaxis warranted in patients who remain at risk of late clinically significant cytomegalovirus infection.

therapy [PET] for viraemia, onset of end-organ disease, or both) is during the first 100 days following HSCT. However, a considerable risk of cytomegalovirus reactivation persists beyond these 100 days (ie, for late cytomegalovirus infection and disease) in some patients.

Letermovir, a cytomegalovirus terminase complex inhibitor, was evaluated in a large, global, phase 3 registrational study (the P001 study)[4] for its safety and efficacy when used for prophylaxis against cytomegalovirus infection in cytomegalovirus-seropositive adult recipients of an allogeneic HSCT. Letermovir was well tolerated in this study and was shown to be superior to placebo in reducing the incidence of clinically significant cytomegalovirus infection until week 24 (around 200 days) after HSCT, when administered up to week 14 (around 100 days).[4] Based on the results of this study, letermovir has been approved in more than 68 countries worldwide for prophylaxis against cytomegalovirus infection and disease in cytomegalovirus-seropositive adult recipients of HSCT.

In the P001 study, an increased incidence of clinically significant cytomegalovirus infection (approximately 12·1%) between 100 days and 200 days following HSCT was observed when letermovir administration was discontinued at around 100 days (unpublished). Similar observations were made in several single-centre retrospective studies, in which the incidence of clinically significant cytomegalovirus infection following letermovir discontinuation at day 100 following HSCT was between 5% and 20%.[5–8] Post-hoc analyses indicated that graft-versus-host disease (GVHD), concomitant corticosteroid use, and baseline high-risk stratum (as defined in the P001 study[4]) were associated with development of clinically significant cytomegalovirus infection following completion of 100 days of letermovir.

We aimed to evaluate the efficacy and safety of extending the duration of letermovir prophylaxis from 100 days to 200 days following HSCT in cytomegalovirus-seropositive recipients of allogeneic HSCT who remained at high risk of clinically significant cytomegalovirus infection 100 days following transplantation.

## Methods

### Study design and participants

In this multicentre, randomised, double-blind, placebo-controlled, phase 3 trial, participants were recruited from 32 hospitals and medical centres across the following six countries: France, Germany, Italy, Japan, the UK, and the USA (appendix pp 2–3). All potential study participants were screened for inclusion and exclusion criteria within 14 days before randomisation.

See Online for appendix

Eligible participants were cytomegalovirus-seropositive adult (aged ≥18 years) recipients of allogeneic HSCT within the 100 days before randomisation, had an undetectable quantity of cytomegalovirus DNA in plasma samples collected within the 14 days before randomisation, had received letermovir as primary prophylaxis initiated within 28 days of HSCT and continued for up to 100 days (93–107 days) following HSCT before randomisation, and were at high risk of clinically significant cytomegalovirus infection. High risk of cytomegalovirus infection, disease, or both was defined as meeting one or more of the following criteria: having a related donor with at least one mismatch at one of the three specified HLA gene loci (*HLA-A*, *HLA-B*, or *HLA-DR*); having an unrelated donor with at least one mismatch at one of the four specified HLA gene loci (*HLA-A*, *HLA-B*, *HLA-C*, or *HLA-DRB1*); having a haploidentical donor; having umbilical cord blood as the stem-cell source; recipients of ex-vivo grafts depleted of T cells, anti-thymocyte globulin, or alemtuzumab; or having GVHD or other conditions requiring the use of systemic prednisone (or its equivalent) at a dose of at least 1 mg/kg of bodyweight per day within 6 weeks of randomisation.

Patients were excluded if they reported severe liver impairment, end-stage renal impairment with an estimated creatinine clearance of up to 10 mL/min, current or recent (within the 7 days before screening) treatment with antiviral agents with anti-cytomegalovirus activity (including acyclovir, valacyclovir, and famciclovir at doses above those recommended for prophylaxis against herpes simplex virus and varicella zoster virus), a history of cytomegalovirus end-organ disease, receiving PET for cytomegalovirus before randomisation, or a history of over 14 days of letermovir interruption during the first 100 days after HSCT before randomisation. In addition, patients who were concurrently participating in, had previously participated in, or planned to participate in any other study of a cytomegalovirus vaccine or cytomegalovirus investigational agent during this study were excluded. Full details regarding the eligibility criteria can be found in the appendix (pp 4–5).

This trial was conducted in accordance with the principles of Good Clinical Practice and was approved by the appropriate institutional review boards and regulatory agencies. All participants provided written informed consent.

### Randomisation and masking

After receiving letermovir for 100 days following HSCT, eligible participants were randomly assigned (2:1) to receive either an additional 100 days of letermovir (ie, letermovir group) or 100 days of a placebo comparator (ie, placebo group). Randomisation was done with a central interactive response technology system, stratified by study centre and haploidentical donor (yes or no).

Double-blinding with in-house masking was used. Participants, investigators, and sponsor personnel or delegates involved in the study intervention administration or clinical evaluation of the participants were masked to the intervention assignments. There were no premature unmasking events. Additional details are provided in the protocol (appendix pp 53–54).

### Procedures

All study participants who had received letermovir for 100 days (around week 14) following HSCT were assigned to receive either letermovir 480 mg once daily (adjusted to 240 mg once daily for participants receiving cyclosporin A) in the letermovir group or a placebo comparator (placebo group) administered orally or intravenously until 200 days (around week 28) after HSCT. Full details of letermovir administration can be found in the appendix (p 5).

If cyclosporin A was initiated after starting the study intervention, the next dose of study intervention was decreased to 240 mg once daily. If cyclosporin A was discontinued permanently or for the long term in a participant already receiving the study intervention, the next dose of study intervention was increased to 480 mg once daily. If cyclosporin A was temporarily withheld because of high concentrations detected by therapeutic blood monitoring, the dose of study intervention did not require dose adjustment.

To assess efficacy, cytomegalovirus DNA viral load was measured every 2 weeks from week 14 to week 40 and every 4 weeks thereafter until week 48, as well as at the cytomegalovirus infection visit or at the early discontinuation visit. Samples were sent to the central laboratory, where cytomegalovirus DNA PCR testing was performed by use of a quantitative cytomegalovirus DNA PCR assay (COBAS AmpliPrep/COBAS TaqMan assay; Roche, CA, USA). Clinical chemistry and haematology were performed at screening and every 2 weeks until week 30. Coagulation (prothrombin time and international normalised ratio) was performed at screening and every 2 weeks until week 28. Urinalysis was performed at screening and at week 28. Pregnancy testing was performed on serum samples (β-human chorionic gonadotropin) at screening and on urine samples at weeks 14, 18, 22, and 26 from women of childbearing potential. HIV, hepatitis B, and hepatitis C tests were performed at screening. Additional details can be found in the protocol (appendix pp 29–33).

Monitoring of adverse events was performed at screening and every 2 weeks during the treatment period (weeks 14–28), then every 4 weeks until week 48. From the time of treatment allocation to 14 days following cessation of treatment, all adverse events, serious adverse events (SAEs), and other reportable safety events were reported by the site investigator. Thereafter, only SAEs considered to be drug-related or leading to death were reported until week 48 after HSCT. Types of adverse events and other reportable safety events included non-serious adverse events, SAEs, pregnancy or lactation

exposure, cancer, overdose, and events of clinical interest that might or might not require regulatory reporting.

### Outcomes

The primary efficacy endpoint was the proportion of participants with clinically significant cytomegalovirus infection from randomisation at week 14 (approximately 100 days following HSCT) to the end of prophylaxis at week 28 (200 days following HSCT). Clinically significant cytomegalovirus infection was defined as the initiation of PET for documented cytomegalovirus viraemia, the onset of cytomegalovirus end-organ disease, or both. Although the protocol recommended that investigators consider initiating PET when cytomegalovirus DNA viral load was above approximately 300 copies per mL, this threshold was provided as guidance and was not mandated. The threshold for initiating PET was left to the clinical judgment of the investigator, given that there is currently no consensus regarding this issue and institutional practice varies widely.

All cases of investigator-reported cytomegalovirus disease were evaluated and confirmed by an independent, external, masked clinical adjudication committee, which used the definitions published by Ljungman and colleagues.[9] The committee reviewed clinical, virological, and histopathological data, as well as the investigators' assessments, for all potential cases of cytomegalovirus disease.

Secondary efficacy endpoints included the proportion of participants with clinically significant cytomegalovirus infection from randomisation at week 14 to week 38 and to week 48; time to onset of clinically significant cytomegalovirus infection from week 14 to week 28 and to week 48; the proportion of participants with PET for cytomegalovirus viraemia from week 14 to week 28 and to week 48; the proportion of participants with all-cause mortality from week 14 to week 28 and to week 48; and time to all-cause mortality from week 14 to week 28 and to week 48.

Safety and tolerability were primarily evaluated by the reporting of adverse events and discontinuation of treatment due to adverse events. All adverse events were reported during the treatment phase, which extended from the day of study entry to 14 days after the last dose of letermovir. Thereafter, only drug-related SAEs or SAEs leading to death were reported until study completion.

To supplement routine safety monitoring, periodic safety reviews were performed by an external data monitoring committee approximately every 6 months to make recommendations for the discontinuation of the study or protocol modifications. No formal interim analyses for efficacy were planned for this study.

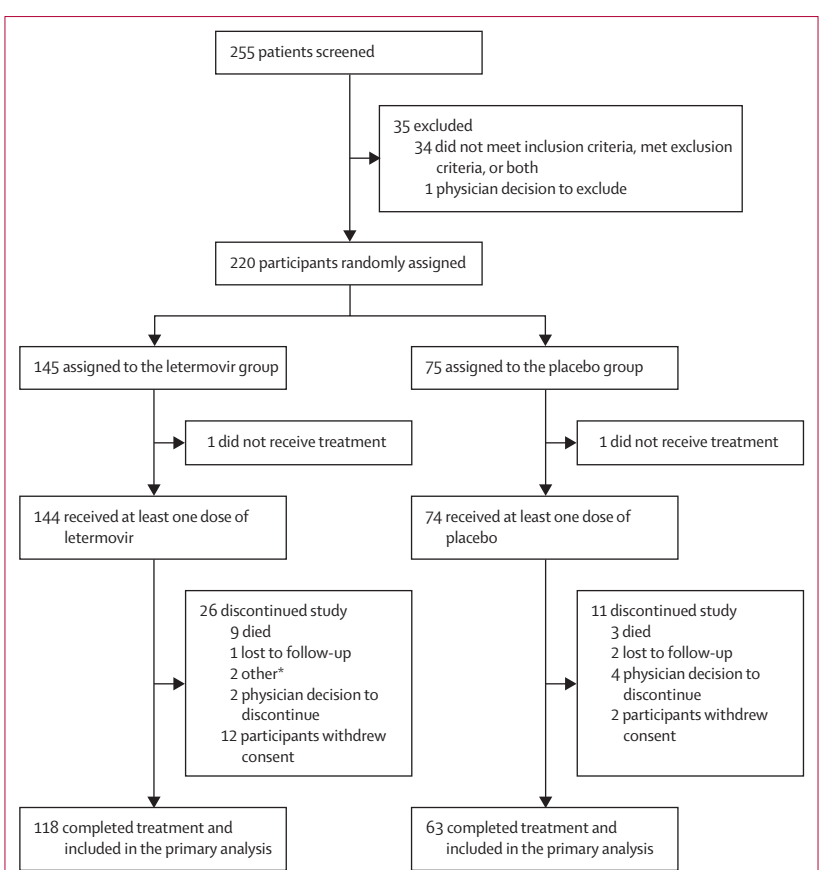


***Figure 1:*** **Trial profile**
*One participant was discontinued because of COVID-19 and one participant had compliance issues and ultimately withdrew consent.

### Statistical analysis

The primary study hypothesis was that letermovir was superior to placebo in the prevention of clinically significant cytomegalovirus infection when letermovir prophylaxis was extended from 100 days to 200 days following HSCT. The planned sample size was 216, which had 80% power at an overall one-sided 2·5% alpha level to establish the primary objective. The sample size calculations assumed that the incidence of clinically significant cytomegalovirus infection would be 8% for the letermovir group and 22% for the placebo group; these rates were estimated with data from participants in the high-risk stratum from the P001 study.[4]

We calculated 95% CIs and one-sided p values for treatment differences in the percentage response using the Mantel-Haenszel method, with continuity correction and stratification according to haploidentical donor (yes or no), to test the superiority of the intervention over placebo for the prevention of clinically significant cytomegalovirus infection from 100 days to 200 days following HSCT. A one-sided p value (<0·025) was used to declare statistical significance of the primary efficacy endpoint. A nominal p value was provided to assess the strength of evidence for the secondary efficacy endpoints.

We estimated the time to onset of clinically significant cytomegalovirus infection using the non-parametric Kaplan-Meier method. The Kaplan-Meier curve was

plotted by treatment group and a nominal p value for the difference between groups in time to onset of clinically significant cytomegalovirus infection was provided by use of the stratified log-rank test stratified by haploidentical donor (yes or no).

We estimated differences in adverse events and their corresponding 95% CIs using Miettinen and Nurminen's method.[10] Safety endpoints were the broad adverse event categories of the proportion of participants with any adverse events, drug-related adverse events, SAEs, drug-related SAEs, or treatment discontinuation due to adverse events, as well as individual adverse events reported by at least eight participants in the letermovir group and at least two participants in the placebo group.

The primary efficacy population was the full analysis set, defined as all randomly assigned participants who received at least one dose of the study intervention. The observed failure approach for handling missing data values was used for efficacy analyses. In this approach, failure was defined as all participants who developed clinically significant cytomegalovirus infection or discontinued the study prematurely with cytomegalovirus viraemia between week 14 and week 28. Safety was analysed in all participants as treated, defined as all enrolled participants who received at least one dose of study drug according to the intervention they were assigned to. Participants who did not receive at least one dose of study intervention were excluded from further analyses.

All statistical analyses were performed with SAS (version 9.4). This study is registered with ClinicalTrials.gov, NCT03930615.

### Role of the funding source

The funder provided letermovir and was involved in study design, data collection, data analysis, data interpretation, writing of the report, and approval for publication. The funder provided financial support for medical writing.

## Results

Between June 21, 2019, and March 16, 2022, 255 patients were screened for eligibility, of whom 220 (86%) participants were randomly assigned to either the letermovir group (145 [66%]) or the placebo group (75 [34%]). Overall, 218 participants were treated with at least one dose of the study intervention and 181 completed the study (figure 1). The most common reasons for study discontinuation overall were withdrawal of consent (14 participants [6%]) and death (12 [6%]). Of the 12 (8%) participants who withdrew consent in the letermovir group, six (50%) reported a desire to not continue participating in the study, four (33%) reported transportation or logistical issues, and two (17%) discontinued treatment because of adverse events (one due to nausea and one due to vomiting). Both of these adverse events are consistent with the known adverse event profile of letermovir. In the placebo group, two (3%) participants withdrew consent

| | Letermovir group (n=144) | Placebo group (n=74) |
|---|---|---|
| Sex | | |
| Female | 52 (36%) | 31 (42%) |
| Male | 92 (64%) | 43 (58%) |
| Age, years | 55 (22–74) | 55 (20–74) |
| Race | | |
| White | 113 (79%) | 60 (81%) |
| Asian | 16 (11%) | 8 (11%) |
| Other | 7 (5%) | 2 (3%) |
| Data missing* | 8 (6%) | 4 (5%) |
| Region† | | |
| Asia-Pacific | 13 (9%) | 4 (5%) |
| Europe | 98 (68%) | 52 (70%) |
| North America | 33 (23%) | 18 (24%) |
| Primary reason for HSCT | | |
| Acute myeloid leukaemia | 61 (42%) | 30 (41%) |
| Acute lymphocytic leukaemia | 23 (16%) | 9 (12%) |
| Myelodysplastic syndrome | 17 (12%) | 6 (8%) |
| Lymphoma | 8 (6%) | 9 (12%) |
| Myelofibrosis | 8 (6%) | 5 (7%) |
| Other disease | 27 (19%) | 15 (20%) |
| Cytomegalovirus-seropositive donor | 86 (60%) | 56 (76%) |
| Cytomegalovirus-seropositive recipient | 143 (99%)‡ | 74 (100%) |
| HLA matching and donor type | | |
| Matched related | 17 (12%) | 11 (15%) |
| Mismatched related | 48 (33%) | 23 (31%) |
| Matched unrelated | 37 (26%) | 23 (31%) |
| Mismatched unrelated | 42 (29%) | 17 (23%) |
| Haploidentical donor | 45 (31%) | 22 (30%) |
| Stem-cell source | | |
| Peripheral blood | 117 (81%) | 62 (84%) |
| Bone marrow | 19 (13%) | 7 (10%) |
| Umbilical cord blood | 8 (6%) | 5 (7%) |
| Conditioning regimen | | |
| Myeloablative | 73 (51%) | 33 (45%) |
| Reduced intensity | 46 (32%) | 27 (37%) |
| Non-myeloablative | 25 (17%) | 14 (19%) |
| Undetectable cytomegalovirus on day of randomisation | 139 (97%) | 71 (96%) |
| GVHD at study entry | | |
| Acute | 25 (17%) | 9 (12%) |
| Chronic | 3 (2%) | 1 (1%) |
| Acute and chronic | 2 (1%) | 0 |
| None | 114 (79%) | 64 (87%) |
| Cytomegalovirus risk factors§ | | |
| Receipt of anti-thymocyte globulin | 67 (47%) | 35 (47%) |
| Receipt of alemtuzumab | 13 (9%) | 9 (12%) |
| Use of ex-vivo grafts depleted of T cells | 15 (10%) | 7 (10%) |

Data are n (%) or median (range). All participants received 100 days of letermovir after transplantation before entering the study. Thereafter, they either received an additional 100 days of letermovir (ie, letermovir group) or 100 days of placebo (ie, placebo group). GVHD=graft-versus-host disease. HSCT=haematopoietic stem cell transplantation. *Data missing when participant did not know race or chose not to report race because of local regulations. †Countries include France, Germany, Italy, Japan, the UK, and the USA. ‡One participant was a cytomegalovirus-seronegative recipient. This individual was not excluded from any analyses. §A comprehensive list of the risk factors evaluated in this study is provided in the appendix (p 6).

***Table 1:*** **Baseline participant characteristics**

because of the participants' desire to not continue participating in the study.

Participant characteristics at study entry were generally similar between both treatment groups (table 1). Most participants were male, White, and European. The proportion of cytomegalovirus-seropositive donors was higher in the placebo group (56 [76%]) than in the letermovir group (86 [60%]). The most common primary reasons for HSCT were acute myeloid leukaemia, acute lymphoblastic leukaemia, and myelodysplastic syndrome. The proportion of participants with GVHD at study entry was higher in the letermovir group than in the placebo group.

Risk factors among participants for late cytomegalovirus infection, disease, or both were balanced between treatment groups; approximately two thirds of participants had two or more risk factors (appendix p 6). Among all 218 participants who received at least one dose of the assigned treatment, the most common risk factors were receipt of anti-thymocyte globulin (102 [47%]); having an HLA-related donor with at least one mismatch at *HLA-A*, *HLA-B*, or *HLA-DR* (71 [33%]); and having a haploidentical donor (67 [31%]).

The median duration of administration of the study intervention was 98 days (range 9–109) in the letermovir group and 97 days (18–106) in the placebo group. Of the 220 participants randomly assigned, one (<1%) participant in the letermovir group and one (1%) participant in the placebo group did not receive a single dose of study intervention and were excluded from all further analyses. The remaining 218 participants who received at least one dose of study intervention were included in both the primary efficacy analysis population and the safety analysis population.

By use of the observed failure approach for missing data, the proportion of participants with clinically significant cytomegalovirus infection from baseline (week 14) to week 28 (200 days following HSCT) was lower in the letermovir group than in the placebo group (four [3%] of 144 *vs* 14 [19%] of 74; treatment difference –16·1% [95% CI –25·8 to –6·5]; one-sided p=0·0005; table 2). All sensitivity analyses accounting for potentially confounding factors showed the superiority of extending the duration of letermovir prophylaxis to 200 days compared with 100 days for the prevention of clinically significant cytomegalovirus infection, which was consistent with the primary efficacy endpoint analysis (appendix p 7).

19 (13%) w from both baseline to week 38 and from baseline to week 48 (one-sided p=0·16; table 2). The time to onset of clinically significant cytomegalovirus infection was substantially delayed in the letermovir group compared with the placebo group from baseline to week 28 (nominal one-sided p<0·0001; figure 2). The time to onset of clinically significant cytomegalovirus infection from baseline to week 48 was similar between both treatment groups (nominal p=0·14; figure 2).

By use of the observed failure approach, the proportion of participants who initiated PET for cytomegalovirus viraemia between baseline and week 28 was substantially lower in the letermovir group (three [2%]) than in the placebo group (12 [16%]; nominal p=0·0012; table 2). The observed failure approach included those who prematurely discontinued from the study with cytomegalovirus viraemia in the failure category. All-cause mortality was similar between both treatment groups from baseline to week 28 and from baseline to week 48 (table 2). The time to all-cause mortality from week 14 to week 28 and week 48 following HSCT was similar between treatment groups (appendix pp 11–12).

Overall, both treatment groups showed a similar adverse event profile. No imbalances were identified

| | Letermovir group (n=144) | Placebo group (n=74) | Treatment difference (95% CI)* | p value |
|---|---|---|---|---|
| **Primary endpoint†** | | | | |
| Failures‡ from week 14 to week 28 | 4 (3%) | 14 (19%) | −16·1 (−25·8 to −6·5) | 0·0005 |
| Clinically significant cytomegalovirus infection | 2 (1%) | 13 (18%) | .. | .. |
| Initiation of PET based on documented cytomegalovirus viraemia | 1 (<1%) | 11 (15%) | .. | .. |
| Onset of end-organ disease | 1 (<1%) | 2 (3%) | .. | .. |
| Discontinued from study with cytomegalovirus viraemia before week 28 | 2 (1%) | 1 (1%) | .. | .. |
| **Key secondary endpoints** | | | | |
| Clinically significant cytomegalovirus infection§ | | | | |
| From week 14 to week 38 | 19 (13%) | 14 (19%) | −5·7 (−16·8 to 5·4) | 0·16 |
| From week 14 to week 48 | 19 (13%) | 14 (19%) | −5·7 (−16·8 to 5·4) | 0·16 |
| Failures¶ from week 14 to week 28 | 3 (2%) | 12 (16%) | −14·1 (−23·3 to −5·0) | 0·0012 |
| Initiation of PET based on documented cytomegalovirus viraemia | 1 (<1%) | 11 (15%) | .. | .. |
| Discontinued from study with cytomegalovirus viraemia before week 28 | 2 (1%) | 1 (1%) | .. | .. |
| All-cause mortality | | | | |
| From week 14 to week 28 | 3 (2%) | 1 (1%) | 0·7 (−3·8 to 5·3) | 0·62 |
| From week 14 to week 48 | 12 (8%) | 6 (8%) | 0·3 (−7·9 to 8·4) | 0·53 |

Data are n (%), unless otherwise indicated. PET=pre-emptive therapy. *Calculated with a stratum-adjusted Mantel-Haenszel method, with the difference weighted by the harmonic mean of sample size per group for each stratum (high or low risk). A one-sided p value (≤0·025) was used to declare significance in the primary analysis. Nominal one-sided p values (not adjusted for multiplicity) are provided for other analyses as a measure of the strength of the relationship between treatment and response. †The proportion of participants who developed clinically significant cytomegalovirus infection or discontinued the study prematurely with cytomegalovirus viraemia from week 14 to week 28 in the primary efficacy population (ie, participants who received at least one dose of study intervention). ‡For the primary endpoint, an observed failure approach was used to handle missing data values, in which failure was defined as all participants who developed clinically significant cytomegalovirus infection or discontinued prematurely from the study with cytomegalovirus viraemia; the categories of failure are mutually exclusive and based on the hierarchy of categories in the order listed. §Defined as the onset of cytomegalovirus end-organ disease (proven or probable) or initiation of PET based on documented cytomegalovirus viraemia and the clinical condition of the participant. ¶For the secondary endpoint, an observed failure approach was used to handle missing data values, in which failure was defined as all participants who initiated PET based on documented CMV viremia or discontinued prematurely from the study with cytomegalovirus viraemia; the categories of failure are mutually exclusive and based on the hierarchy of categories in the order listed.

***Table 2:*** **Efficacy endpoints in the primary efficacy population**

between treatment groups in the broad categories of adverse events during the treatment phase (appendix p 8). The most commonly reported adverse events during the treatment phase were new-onset GVHD, nausea, diarrhoea, pyrexia, and decreased appetite (table 3; appendix p 9). The frequency of adverse events related to blood and lymphatic system disorders were similar between the letermovir group (23 [16%]) and the placebo group (11 [15%]); none were considered to be drug-related by investigators or led to discontinuation of either study intervention (data not shown). No participant received a dose reduction because of reported adverse events. No participant discontinued either treatment because of drug-related toxicity. There were no treatment-related deaths.

The number of participants with acute myeloid leukaemia relapse was higher in the letermovir group (nine [6%]) than in the placebo group (one [1%]; appendix p 10); however, this difference was not statistically significant (data not shown). In the letermovir group, seven (5%) participants discontinued study treatment because of the following adverse events: dyspepsia, pyrexia, GVHD, COVID-19, lymph node tuberculosis, recurrent acute lymphocytic leukaemia, and post-transplantation lymphoproliferative disorder. In the placebo group, one (1%) participant discontinued treatment because of GVHD.

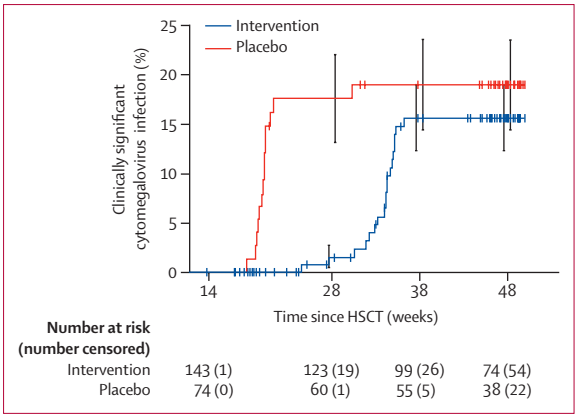


***Figure 2:*** **Cumulative rate of clinically significant cytomegalovirus infection in the primary efficacy population**
Kaplan-Meier plot for the time to onset of clinically significant cytomegalovirus infection from randomisation at week 14 to week 48 following HSCT. HSCT=haematopoietic stem-cell transplantation.

| | Letermovir group (n=144) | Placebo group (n=74) | Difference (95% CI)* |
|---|---|---|---|
| Any adverse event | 128 (89%) | 69 (93%) | −4·4 (−11·8 to 4·7) |
| GVHD | 43 (30%) | 23 (31%) | −1·2 (−14·5 to 11·2) |
| Diarrhoea | 17 (12%) | 9 (12%) | −0·4 (−10·7 to 8·2) |
| Nausea | 16 (11%) | 13 (18%) | −6·5 (−17·6 to 2·9) |
| Pyrexia | 13 (9%) | 9 (12%) | −3·1 (−13·3 to 5·0) |
| Decreased appetite | 6 (4%) | 9 (12%) | −8·0 (−17·8 to −0·8) |

Data are n (%), unless otherwise indicated. Most common adverse events defined as adverse events of any severity that were reported in at least 10% of participants in either treatment group. Information on AE grades was not collected in this trial. GVHD=graft-versus-host disease. *Based on Miettinen and Nurminen's method.[10]

***Table 3:*** **Adverse events in the safety population**

## Discussion

Although the risk of cytomegalovirus reactivation decreases after the first 100 days following HSCT as reconstitution of the immune system occurs and immunosuppressant dosages are decreased, a subgroup of HSCT recipients remain at risk of late cytomegalovirus infection or disease. Factors associated with continued risk of reactivation include the type of transplantation (eg, haploidentical or cord blood), the degree of mismatch and relatedness between donor and recipient, the use of grafts depleted of T cells, and post-transplantation complications—eg, new-onset GVHD or other conditions requiring treatment with steroids.[11–14] The standard of care for prevention of clinically significant cytomegalovirus infection or disease after 100 days following HSCT is PET, which is the practice of active surveillance for viral replication with antiviral treatment, started only when cytomegalovirus viraemia is detected.[9]

This study showed that extending the duration of letermovir prophylaxis to 200 days following HSCT is superior to 100 days of letermovir in preventing late clinically significant cytomegalovirus infection in allogeneic HSCT recipients. This superiority was maintained despite the higher proportion of participants with GVHD at study entry in the letermovir group than in the placebo group and 12 participants in the letermovir group withdrawing from the study. 200 days of letermovir prophylaxis was well tolerated and had a similar safety profile to 100 days of letermovir prophylaxis, which is consistent with safety findings reported in the P001 study.[4] In particular, there was no evidence of myelotoxicity associated with letermovir use, which is relevant in the patient population of HSCT recipients.

After discontinuation of letermovir at 200 days (28 weeks) following transplantation, there was an increase in the incidence of clinically significant cytomegalovirus infection until week 38 in the letermovir group, which was similar to the incidence in the placebo group at that timepoint. The rebound in the incidence of clinically significant cytomegalovirus infection after letermovir discontinuation was not entirely unexpected, given that letermovir use results in virological suppression, not cure. There were no additional cases of clinically significant cytomegalovirus infection among participants in either treatment group at week 48, which could reflect a degree of cytomegalovirus-specific immunity adequate to prevent further viral reactivation beyond week 38 following HSCT.

It remains unclear whether there might be additional benefits to extending letermovir prophylaxis beyond 200 days following HSCT. A 2022 systematic literature review and meta-analysis of 48 unique observational

studies, most of which were single-centre, concluded that primary prophylaxis with letermovir was effective in reducing cytomegalovirus-related complications and overall mortality in patients beyond 200 days following transplantation.[15] In the meantime, it would be judicious to continue monitoring cytomegalovirus replication after discontinuation of letermovir to allow for timely initiation of PET with other cytomegalovirus agents if necessary; no cross-resistance with letermovir has been reported to date. The decision to extend letermovir prophylaxis beyond 200 days should be left to the clinical judgment of the treating physician, who should assess the risk versus the benefit of continued virological suppression in the context of the patient's immune system recovery. In this regard, the role of cytomegalovirus-specific cell-mediated immunity in controlling viral replication in the HSCT setting needs further research.

An important controversy regarding cytomegalovirus after allogeneic HSCT is the protective effect of cytomegalovirus replication on relapse of acute leukaemia.[2,16,17] Although a numerical imbalance was observed in the incidence of acute myeloid leukaemia relapse between treatment groups in this trial, this difference was not statistically significant. No trends were observed in the timing of recurrent acute myeloid leukaemia events relative to the treatment duration of letermovir (appendix p 10), and none of the events were considered to be drug-related by the study investigators. Thus, it is unlikely that the recurrent acute myeloid leukaemia events seen in this study were related to the suppression of cytomegalovirus replication by letermovir.

This study has several data gaps, which might limit the generalisability of our conclusions to a larger population of HSCT recipients at risk of late cytomegalovirus infection and disease. For example, although most categories of risk factors for cytomegalovirus reactivation were represented in the study population, data related to some additional risk factors (eg, patients who received cyclophosphamide after HSCT) were not collected systematically. Additionally, we do not have documentation for adherence to letermovir prophylaxis during the first 100 days before study entry, although patients who missed more than 14 days of letermovir in those 100 days were excluded from the study. Furthermore, other than the fact that eligible participants could not have received PET before randomisation and had to have an undetectable or unquantifiable viral load of cytomegalovirus DNA in the 14 days before randomisation, no further information is available on the patterns of detectable viraemia in participants during the pre-study period.

Another study limitation is that a mortality benefit could not be definitively established in this study, which is due to the study design and sample size. Given that participants were randomly assigned after receiving letermovir prophylaxis for 100 days following HSCT, the design did not allow for accounting of deaths or cases of clinically significant cytomegalovirus infection that had occurred before study entry. The study entry criteria also precluded enrolment of participants with clinically significant comorbidities and participants had to be clinically stable and reasonably adherent to primary prophylaxis with letermovir before study entry, which would have further decreased the risk of mortality in the study population. Thus, patients with the highest risk of cytomegalovirus-related mortality could have been excluded from the study. Emerging real-world data and evidence on extending the duration of letermovir prophylaxis will provide further clarity on several issues, such as additional categories of patients at risk of cytomegalovirus reactivation who might benefit from 200 days of letermovir prophylaxis following HSCT, whether or not extending letermovir prophylaxis beyond 200 days is necessary in a select group of HSCT recipients, and if there is a mortality benefit associated with extending the duration of prophylaxis beyond the first 100 days after HSCT.

Nevertheless, the availability of letermovir for prophylaxis against late cytomegalovirus infection and disease provides transplantation physicians with an attractive new option over PET for the prevention of late cytomegalovirus reactivation in this patient population. Other currently available anti-cytomegalovirus agents (eg, ganciclovir, valganciclovir, foscarnet, and cidofovir) used for PET are associated with clinically significant myelotoxicity, nephrotoxicity, or both; myelotoxicity is particularly relevant when treating HSCT recipients. The safety findings in both this study and in another large phase 3 trial completed in 2023, which evaluated cytomegalovirus prophylaxis with letermovir for 200 days in kidney transplant recipients,[18] substantiate that letermovir is well tolerated and has a favourable safety profile for up to 200 days in the transplantation setting. Furthermore, no resistance associated with extended letermovir use was noted in either study. Additionally, the issue of cross-resistance among other anti-cytomegalovirus antiviral agents limits and complicates management options in instances of PET failure due to the development of antiviral resistance. By contrast, because of its unique mechanism of action, letermovir does not cross-react with other available anti-cytomegalovirus agents, allowing for the use of alternative agents in instances of PET failure. Furthermore, extending viral suppression with letermovir beyond 100 days following transplantation would delay or even obviate the need for PET. This situation would represent an advance in the patient-centric, risk-adapted approach to letermovir prophylaxis in situations where the risk of clinically significant cytomegalovirus infection remains high beyond day 100; for example, with the onset of GVHD and its treatment course, which are ongoing dynamic events following HSCT. In conclusion, extending the duration of letermovir prophylaxis to 200 days following HSCT is

efficacious and safe in reducing the incidence of late clinically significant cytomegalovirus infection in patients at risk.

#### Contributors

VLT, BH, CBo, and CBa designed the study. All authors analysed and interpreted the data. DR, VLT, BH, CBo, and CBa directly accessed and verified the underlying data reported in the manuscript. All authors critically reviewed the manuscript and approved the final version for submission. All authors had full access to all the data in the study and had final responsibility for the decision to submit for publication.

#### Declaration of interests

SSD has received research grants (to his institution) from Allovir, Ansun Biopharma, Karius, and Merck; served as advisory board member or consultant for Allovir, Asepticope, Merck, and Takeda; and served on speakers' bureaus for Astellas Pharma, Merck, and Takeda. MSc has received research grants from Apogenix, Hexal, and Novartis; travel support from Hexal and Kite; financial support for educational activities and conferences from BMS and Kite; compensation as a member of the scientific advisory board of MSD, Novartis, BMS, and Pierre Fabre; and is co-founder and shareholder of ToleroGenixX. MSt holds an advisory role for MSD and has received honoraria from MSD. BH, CBo, and CBa are current employees of Merck Sharp & Dohme LLC, a subsidiary of Merck & Co (Rahway, NJ, USA) and may own stock or stock options in Merck & Co. At the time of the study and during initial phases of manuscript development, VLT was an employee of Merck Sharp & Dohme and may own stock or stock options in Merck & Co. All other authors declare no competing interests.

#### Data sharing

The data sharing policy (including restrictions) of Merck Sharp & Dohme is available at http://engagezone.msd.com/ds_documentation.php. Requests for access to the clinical study data can be submitted through the Engage Zone site or via email to the Data Access mailbox at dataaccess@merck.com.

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