# End of trial report

| | |
|---|---|
| **Sponsor:** | HOVON foundation |
| **Trial:** | HOVON 107 MOBILIZATION |
| **EudraCT:** | 2010-023436-16 |
| **Title of study:** | The feasibility and efficacy of subcutaneous Plerixafor for mobilization of peripheral blood stem cells in allogeneic HLA–identical sibling donors: a prospective phase II study. |
| **Report date:** | 17 June 2020 |

| **Signature** | |
|---|---|
| Name: | Dr. G.E. de Greef |
| Function: | Principal Investigator |
| Date: | 24-jun-20 |
| Signature: | DocuSigned by: Dr. G.E. de Greef 1D23C08F887A4A3... |

**Investigational sites & investigators:**

| Country | City | Organization | Local Investigator |
|---|---|---|---|
| Nederland | Rotterdam | Erasmus MC | mw. Dr. G.E. de Greef |
| Nederland | Amsterdam | VUMC | dhr. Dr. J.J.W.M. Janssen |
| Germany | Leipzig | University of Leipzig | Dr. D. Niederwieser |
| Nederland | Utrecht | UMCU | mw. Dr. E.J. Petersen |

**Publications:**

The feasibility and efficacy of subcutaneous plerixafor for mobilization of peripheral blood stem cells in allogeneic HLA-identical sibling donors: results of the HOVON-107 study.
de Greef GE, Braakman E, van der Holt B, Janssen JJWM, Petersen E, Vucinic V, Thuss N, Grootes M, Cornelissen JJ.
Transfusion. 2019 Jan;59(1):316-324. doi: 10.1111/trf.15037. Epub 2018 Dec 11.

**Study period:**

21May2012 (FPI) – 28Jan2019 (LPLV)

**Phase of development:**

Phase II

**Objectives:**

**Primary Objective:**

♦ To determine the feasibility of plerixafor 320 µg/kg subcutaneously to harvest a sufficient number of peripheral blood stem cells in healthy HLA-matched sibling donors.
Feasibility will be defined as follows: A harvest of at least $2x10^6$ CD34+ cells/kg recipient body weight cells in one or two aphereses in at least 90% of the donors.
A harvest of at least $2x10^6$ CD34+ cells/kg cells in one or two aphereses in 70% or less of the donors will be considered as failure.

**Secondary Objectives:**

**Donors**

♦ To determine the efficacy of plerixafor subcutaneous. Efficacy will be determined as follows:
The absolute number of CD34+ cells collected in 1 liter processed volume.
♦ To determine the time interval that is required to obtain $2.0x\ 10^6$ CD34 + cells from start of mobilization procedure
♦ Pharmacokinetics: to determine the number of CD34+ cells in the peripheral blood at regular intervals after the administration of plerixafor.
♦ To determine the number of CD34+ cells in the peripheral blood as well as in the collected stem cells at regular intervals during the stem cell apheresis.
♦ To determine the phenotype of plerixafor mobilized and collected CD34+ cells including progenitor cells, dendritic cells and regulatory T-cells both in peripheral blood and collected stem cells.
♦ To document adverse events grade 1-4 during and directly following mobilization by plerixafor 320 µg/kg subcutaneously.

**Patients**

♦ To document engraftment 30, 60 and 90 days after transplantation with an allograft harvested after mobilization with plerixafor 320 µg/kg subcutaneous. Engraftment is defined as the first of 3 consecutive days with an absolute neutrophil count (ANC) of at least $0.5 \times 10^9$/l
♦ To document the day of hematopoietic reconstitution for neutrophils and platelets. This is defined as the first of 3 consecutive days of ANC ≥ $0.5x\ 10^6$/L and platelets > $50x10^9$/L
♦ To study chimerism in peripheral blood and T-cells 30, 60, and 90 days, and chimerism in bone marrow 90 days after transplantation with an allograft harvested after mobilization with plerixafor 320 µg/kg subcutaneously.
♦ To evaluate the incidence and grade of graft versus host disease (GVHD).

**Methodology:**

Allogeneic HLA–identical sibling donors received one or two subcutaneous (sc) injections of plerixafor 0.320 mg/kg. The primary endpoint, was defined as feasibility to mobilize a minimum of $2.0\ x10^6$ CD34+ cells/kg recipient weight obtained by leukopheresis in at least 90% of the donors.

**Number of patients:**

Planned: In the initial protocol, 60 allogeneic HLA matched sibling donors and 60 patients were planned. However, due to slow accrual this protocol was amended and the arm 'plerixafor i.v.' was closed.

Enrolled: 38 donors/patients

Enrolled and analyzed: 23 donors/patientsincluded in sc arm

Enrolled but not analyzed, including reason: 14 donors/patients in iv arm

- (only limited analysis was done on this group for supporting information)
- Not evaluable: 1 (no mobilization because patient died)

**Diagnosis and main criteria for inclusion:**

**Donor inclusion criteria**

♦ HLA identical sibling donor

♦ Age 18-60 years inclusive

♦ Hematologic parameters within normal limits

♦ Capable of undergoing leucapheresis: adequate venous access. Must be willing to undergo insertion of a central catheter should leucapheresis via peripheral vein be inadequate

♦ Willing and able to have bone marrow aspiration if there is mobilization failure

♦ Negative pregnancy test at study entry for women of childbearing potential

♦ Willing and able to use adequate contraception during the mobilization period and up to 3 months after last dose of plerixafor

♦ Written informed consent from donor

**Donor exclusion criteria**

♦ Monozygotic twin

♦ Unstable hypertension requiring more than 1 medication.

♦ Positive serology for hepatitis C or HbsAg

♦ Treatment with other investigational drugs

♦ HIV positivity

♦ Any psychological, familial, sociological and geographical condition potentially hampering compliance with the study protocol and follow-up schedule

♦ Pregnant or breastfeeding female subject

**Patient inclusion criteria**

♦ Age 18-65 years inclusive

♦ In general, indication for allogeneic stem cell transplant will be determined by each participating center according to local criteria.

♦ Patients with a cytopathologically confirmed diagnosis of:
- De novo Acute Myeloid Leukemia (AML) according to WHO classification in first complete remission (excluding acute promyelocytic leukemia)
- Myelodysplasia RA(RS)/RCMD with IPSS ≥ 1.5
- Myelodysplasia refractory anemia with excess of blasts (RAEB) with IPSS ≥ 1.5 in first complete remission
- Therapy related AML/RAEB in first complete remission
- De novo B- or T -Lineage Acute Lymphatic Leukemia (ALL) in first complete remission.
- Biphenotypic leukemia in first complete remission
- Multiple Myeloma, not included in other transplant study
- Hodgkin Lymphoma
- Non-Hodgkin Lymphoma
- Chronic Lymfocytic Leukemia (CLL)
- Chronic Myeloid leukemia (CML)

♦ WHO performance score 0,1 or 2

♦ Patients should have an HLA- identical sibling donor

♦ Life expectancy >3 months

♦ Negative pregnancy test at study entry for women of childbearing potential

♦ Willing and able to use adequate contraception

♦ Written informed consent from patient

**Patient exclusion criteria**

♦ Cardiac dysfunction as defined by:
- Myocardial infarction within the last 6 months of study entry
- Reduced left ventricular function with an ejection fraction < 50% as measured by MUGA scan or echocardiogram,
- Unstable angina
- Unstable cardiac arrhythmias

♦ Severe pulmonary dysfunction (CTCAE grade 3-4, see appendix B)

♦ Severe neurological or psychiatric disease

♦ Significant hepatic dysfunction (serum bilirubin ≥ 1.5 times upper limit of normal or transaminases ≥ 2.5 times upper limit of normal)

♦ Significant renal dysfunction (creatinine clearance < 50 ml/min after rehydration)

♦ Concurrent severe and/or uncontrolled medical condition (e.g. uncontrolled diabetes, infection, hypertension, cancer, etc.)

♦ Patient known to be HIV-positive

♦ Pregnant or breast-feeding female patients

♦ Any psychological, familial, sociological and geographical condition potentially hampering compliance with the study protocol and follow-up schedule

♦ Presence of other active non-hematological malignancy

| **Investigational Medicinal Product(s), dose and mode of administration:** |
|---|
| Plerixafor 320 µg/kg, subcutaneous injection at 11 PM, 9 hours before start of stem cell collection.<br>(14 donors/patients were included in arm B of initial protocol and received plerixafor 320 µg/kg, intravenous in 30 minutes, 4 hours before start of the stem cell collection.) |
| **Duration of treatment:** |
| 1 or 2 injections of plerixafor. |
| **Comparator(s), dose and mode of administration:** |
| No comparator. |
| **Duration of treatment:** |
| No comparator. |
| **Criteria for evaluation - Efficacy:** |
| Primary endpoint:<br>- Percentage of donors with a successful harvest. Successful harvest was defined as ≥ $2\times10^6$ CD34+ cells/kg recipient weight after one or two stem cell collections.<br><br>Secondary endpoints for donors:<br>- The absolute number of CD34+ cells collected in 1 liter processed volume;<br>- The time required to collect $2.0\times10^6$ CD34+ cells/kg, also in relation to the time of administration of plerixafor;<br>- The number of CD34+ cells in the peripheral blood at regular intervals after administration of plerixafor;<br>- The number of CD34+ cells in the apheresis product at regular intervals during the stemcell apheresis;<br>- The phenotype of mobilized CD34+ cells including its subpopulations, dendritic cells as well as regulatory T-cells in peripheral blood and in the graft.<br><br>Secondary endpoints for patients:<br>- Incidence of engraftment at days 30, 60 and 90 after transplantation with plerixafor mobilized HPCs. Engraftment is defined by the first of 3 consecutive days with an absolute neutrophil count of at least $0.5\times10^9$/l;<br>- Time to hematopoietic reconstitution. This will be defined as the first of 3 consecutive days of neutrophils ≥ $0.5\text{x }10^9$/L and platelets >$50\text{x}10^9$/L;<br>- Hematopoietic chimerism in blood and CD3 isolated fraction at 30, 60 and 90 days and in bone marrow on day 90 following the allogeneic transplant;<br>- Incidence and grade of acute GVHD |
| **Criteria for evaluation - Safety:** |
| For donors:<br>- The incidence and CTCAE grade (version 4.0) of adverse events.<br>For patients:<br>- Incidence and grade of acute GVHD. |

**Statistical methods:**

The study was originally designed as a randomized, multicenter phase II trial with two treatment arms, i.e., mobilization with either PXF SC (arm A) or IV (arm B).

The aim of the study was to assess the feasibility of each of the two arms separately, as defined before. For each treatment arm, the optimal Simon 2-stage design was applied.

However, a formal comparison between the two arms would not be applied. In each of the two arms, a true percentage of 70% successful harvests would be considered insufficient, while 90% or more would be desirable and warrant further investigation of the arm(s).

With $\alpha = 0.10$ and $\beta = 0.20$, a sample size of 28 patients per arm would be required, with an interim analysis after the first 9 donors. At the final analysis at least 23 of 28 successful had to be observed in one arm to conclude that the arm would be feasible.

Because of slow accrual, the trial was amended to continue only with arm A. The estimated rate of donors with $2.0 \times 10^6$ or more CD34+ cells/kg recipient weight after one or two harvests would be calculated along with the 90% confidence interval (CI).

Continuous variables were summarized by median and range. Adverse events were scored according to the National Cancer Institute's Common Terminology Criteria for Adverse Events, version 4.0, separately for donors and patients.

Progression-free and overall survival rates were estimated by the Kaplan–Meier method, and 95% CIs were constructed. Kaplan–Meier survival curves were generated to illustrate progression-free survival and overall survival.

Subsets of cells were determined. These subsets are described using appropriate statistics (mean+SD, median, range; percentages). For illustrative purposes, these subsets were also obtained in material obtained from donors, age and sex matched, who were previously mobilized with G-CSF. A formal comparison between the two donor groups (plerixafor and G-CSF) however was not performed.

**Summary of efficacy results:**

Results are only reported for arm A (SC plerixafor).

For donors:

- Between August 2012 and January 2017, a total of 24 donors were included in the PXF SC arm. One donor was not treated because the patient had already died.
- 23 donors received PXF 0.320 mg/kg SC, median 9.7 hours (range, 8–10.7), before the planned stem cell collection. In 12 donors, this resulted in 2 × $10^6$/kg or more recipient weight CD34+ cells. In one donor 1.9 × $10^6$/kg CD34+ cells was considered sufficient for transplant, and no second SC injection was performed. Ten of 23 donors needed a second SC injection and stem cell collection. Finally, median 3.3 (1.9–6.5) × $10^6$/kg recipient weight CD34+ cells were collected in all 23 donors and transplanted. These 23 donors and patients were required to consider the SC PXF arm feasible. As the primary endpoint had been achieved, it was decided to close the trial.
- The steady state CD34+ cell number in peripheral blood before the first PXF administration was low, that is, median 3(1–5) x$10^6$ CD34+ cells/L. At 2 hours after SC administration of PXF, an increase of CD34+ cells in blood was apparent: median 13 (5.9–50) x $10^6$/L. At 4 and 6 hours after PXF the CD34+ cell levels were only slightly higher: median 20 (9.6–49) x $10^6$/L and median 20 (11–58)x$10^6$/L, respectively.
- Stem cell collection was performed in 23 donors. The volume of processed blood was median 14.6 (9.1–23.1) L. In 10 donors in which a second collection was performed, a median of 13.4 (7.0–23.1) L of blood was processed. One donor needed an inguinal central catheter due to clotting during the first procedure.
- At the start of stem cell collection, 8 to 11 hours after the administration of PXF, CD34+ cells in the peripheral blood were median 26.1(8.5–71.1)x$10^6$/L. During the apheresis procedure, the numbers of CD34+ cells in the peripheral blood initially dropped but did not change much thereafter.
- After 5 L of processed blood volume, the CD34+ cells were median 18(10–52) x$10^6$/L; after 10 L, 16(6.9–46) x $10^6$/L; and after 15 L, median 15.9(5.7–47.5) x$10^6$/L. The amount of CD34+ cells in the grafts after 5, 10, and 15 L of processed blood volume were median 0.6 (0.2–2.6), 1.4 (0.6–3.4), and 2.1 (1.1–6.6)x$10^6$/kg receiver weight, respectively.
- The grafts obtained after a single SC injection of PXF contained a median of 194 (79–548) x$10^6$ CD34+ cells. Within the CD34+ cell population, 31% showed the phenotype of the primitive hematopoietic stem cell: (lin- CD34+/CD38low/ CD45RA-/CD90+) and 55% had the phenotype of the more committed multipotent/common myeloid progenitor cells (CD34+/CD45RA+/CD90–). A small percentage of CD34+ cells coexpressed CD19 (2.85%).
- The 10 G-CSF mobilized grafts contained a median of 438 x$10^6$ (356–840) CD34+ cells, of which 15% were phenotypic hematopoietic stem cells and 77% phenotypic multipotent/common myeloid progenitor cells. The fraction of CD34+/CD19+ cells was 2%.
- Grafts obtained after PXF mobilization contained more CD3+ cells: median 22.6(10.6–56.7) $10^9$ compared to G-CSF (CD3+ cells: median 12.8(7.6–21) x$10^9$). The CD4/CD8 ratios were comparable, as well as the frequencies of IL-4+/ CD4+ Th2 cells; IL-17+/CD4+/Th17 cells and CD4+ /Foxp3+ regulatory T-cells. Also, the numbers of CD3–/CD16 +/CD56+ NK cells were similar both after PXF and G-CSF. The number of CD19+ B cells in the PXF-mobilized grafts appeared twice as high as in G-CSF-mobilized grafts.

For patients:

- All 23 patients were directly transplanted with the PXF mobilized grafts. Conditioning was myeloablative in 6 patients and non-myeloablative in 17 patients.
- GvHD prophylaxis was given according to local protocol. One patient received cyclophosphamide following transplantation.
- All patients engrafted and recovered, with ANC greater than $0.5x10^9$/L median on Day 17 (0–27) and platelets greater than $50x10^9$/L on Day 13 (0–23);
- Numbers of CD3+ cells, CD3+ /CD4+ cells and CD19+ cells showed a gradual increase and were within normal range at 12 months.
- With respect to chimerism at 3 months, 11 of 16 patients with available data showed full donor chimerism in the peripheral blood. For bone marrow and CD3, this was found in 13 of 15 and 5 of 14 patients. At 12 months, all analyzed patients had complete donor hematopoiesis in all fractions. Data on immunologic reconstitution of 16 of 23 transplant recipients showed CD3/CD8 cells and NK cells at normal range 3 months after transplantation.
- Overall survival and progression-free survival at 12 months were 72% (95% CI, 49%–87%) and 55% (95% CI, 32%–73%). Eight of 23 patients died: 5 because of disease progression, 2 with infection, and 1 with GvHD. Median follow-up of patients still alive is 24 months (6–26).

**Summary of safety results:**

Results are only reported for arm A (SC plerixafor).

For donors:

- Spleen sizes in 23 donors determined by ultrasound were found to be within normal range at the start: median 11 (10–12) cm, and within similar range the day after PXF: median 11 (10–12) cm.
- Side effects related to PXF occurred in 15 of 23 (61%) donors with the majority Grade 1 or 2. CTCAE Grade 3 side effects were clotting during the leukapheresis procedure in two donors, decrease of platelets in one donor, and fatigue after the mobilization procedure in one donor. Grade 3 hypokalemia and syncope were observed each in one donor but considered not related. All side effects resolved. No Grade 4 adverse effects were reported.

For patients:

- Acute GvHD was observed in 6 of 23 patients: 2 patients, Grade 3; 1 patient, Grade 2; and 3 patients, grade 1. One patient died of steroid-refractory GvHD. Chronic extensive GvHD was present in 7 of 23 patients, and limited GvHD in 3 of 23. The latter involved liver and mouth in all patients, skin in 3, eyes in 2, and lungs in 1 patient. At 12 months, it was ongoing in 4 patients.

**Conclusions:**

Stem cell mobilization with 1-2 injections of sc PFX 0.320 mg/kg in allogeneic HLA identical sibling donors is feasible. The limited toxicity of PFX and the need for a restricted number (1 or 2) of injections is an advantage compared to mobilization with multiple injections of G-CSF. Still further studies are needed to confirm the benefit and safety of PFX in comparison to G-CSF in allogeneic stem cell donors. The study showed that PFX mobilizes the most primitive phenotypic stem cells. Engraftment occurred within 30 days both for neutrophils and platelets. In spite of relative high numbers of CD3+ cells, the incidence and severity of acute and chronic GvHD in the patients was not increased.