CLINICAL STUDY PROTOCOL

AN INTERNATIONAL, MULTICENTRE, OPEN-LABEL STUDY TO EVALUATE THE EFFICACY AND SAFETY OF TWO DIFFERENT VACCINATION REGIMENS OF IMMUNOTHERAPY WITH ALLOGENEIC DENDRITIC CELLS, DCP-001, IN PATIENTS WITH ACUTE MYELOID LEUKAEMIA THAT ARE IN REMISSION WITH PERSISTENT MRD

Investigational Product: DCP-001

EudraCT No.: 2017-003426-32

Phase: IIb, Proof of Concept

Protocol Number: DCOne-002

Study Acronym: ADVANCE II
(Allogeneic Dendritic cell Vaccination in AML oNgoing Clinical Evaluation II)

Indication: Acute Myeloid Leukaemia

Authors: A. A. van de Loosdrecht, J.P. Rovers

Protocol version and Dates

Original protocol	V1, 15 August 2017
Amendment 1:	V1.1, 6 October 2017
Amendment 2:	V1.2, 21 November 2017
Amendment 3:	V2.0, 27 August 2018

Client Company:

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1 SPONSORS SIGNATURES

AN INTERNATIONAL, MULTICENTRE, OPEN-LABEL STUDY TO EVALUATE THE EFFICACY AND SAFETY OF TWO DIFFERENT VACCINATION REGIMENS OF IMMUNOTHERAPY WITH ALLOGENEIC DENDRITIC CELLS, DCP-001, IN PATIENTS WITH ACUTE MYELOID LEUKAEMIA THAT ARE IN REMISSION WITH PERSISTENT MRD

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2 PRINCIPAL INVESTIGATOR SIGNATURE

The information contained in this document is CONFIDENTIAL and, except to the extent necessary to obtain informed consent, may not be disclosed unless such disclosure is required by government regulation or state/local customs or law. Persons to whom the information is disclosed must be informed that the information is CONFIDENTIAL and not be further disclosed by them.

By my signature below I agree to conduct this clinical trial in accordance with Good Clinical Practice, the Declaration of Helsinki, government regulations and state/local customs or laws, including those applying to institutional/ethics review and lnformed Consent. I have read the lnvestigator's Brochure and clinical study protocol. I agree to ensure the confidentiality of my patients; however I agree to make available to the Sponsor, the applicable of this clinical study, and relevant regulatory authorities, my patients' medical records. I am aware of my responsibilities as investigator.

Approval Signature

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3 LOCAL INVESTIGATOR SIGNATURE

By my signature below I agree to personally supervise the conduct of this study in my affiliation and to ensure its conduct in compliance with the protocol, Good Clinical Practice, the Declaration of Helsinki, government regulations and state/local customs or laws, including those applying to institutional/ethics review and informed consent. I have read the lnvestigator's Brochure and clinical study protocol. I agree to ensure the confidentiality of my patients; however I agree to make available to the Sponsor, the applicable of this clinical study, and relevant regulatory authorities, my patients' medical records. I am aware of my responsibilities as investigator.

Local Site Name; _____
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Local Investigator Signature: _____ Date_____

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5 LIST OF ABBREVIAITIONS

ADR	Adverse Drug Reaction
AE	Adverse Event
AF	Alkaline phoshatase
ALT	Alanine Amine Transferase
AML	Acute Myeloid Leukemia
ANC	Absolute Neutrophil Count
APL	Acute Promyelocytic Leukemia
APTT	Activated Partial Thromboplastin Time
AST	Aspartate Amino Transferase
BM	Bone Marrow
CFR	Code of Federal Regulations
CHF	Congestive Heart Failure
CHMP	Committee for Medicinal Products for Human Use
CIMT	Association for Cancer Immunotherapy
CMMI	Centre for Microscopy and Molecular Imaging
COA	Certificate of Analysis
CR	Complete Remission
CRA	Clinical Research Associate
CRF	Case Report Form
CRO	Clinical Research Organisation
CTA	Clinical Trial Agreement
CTCAE	Common Terminology Criteria for Adverse Events
CTL	Cytotoxic T-lymphocyte
CVA	Cerebrovascular Accident
DC	Dendritic Cells
DSMB	Data Safety Monitoring Board
DTH	Delayed Type Hypersensitivity
ECG	Electrocardiogram
ECOG	East Cooperative Oncology Group
EDC	Electronic Data Capture
EMA	European Medical Agency
EOS	End Of Study
EOT	End Of Treatment
FDA	US Food and Drug Administration
FU	Follow Up
GCP	Good Clinical Practice
GFR	Glomerular Filtration Rate
GMP	Good Manufacturing Practice

HB	Haemoglobin
HLA	Human Leukocyte Antigen
HOVON	Stichting Hemato-Oncologie voor Volwassenen Nederland
HSCT	Hematopoietic Stem Cell Transplantation
IB	Investigator Brochure
ICH	International Council for Harmonisation
ICS	Intracellular Cytokine Staining
IEC	Institutional Ethical Committee
IFN	Interferon
IHC	ImmunoHistoChemistry
IL	Interleukin
IMP	Investigational Medical Product
IMPD	Investigational Medical Product Dossier
IMV	Interim Monitoring Visit
IP	Investigational Product
ISO	International Organization for Standardization
LAA	Leukaemia Associate Antigen
LAP	Leukaemia Associated Phenotypes
LDH	Lactate DeHydrogenase
LN	Lymph Node
MDRD	Modification of Diet in Renal Disease
MDSC	Myeloid-Derived Suppressor Cells
MFC	Multicolor Flow Cytometry
MHC	Major Histocompatibility Complex
MLR	Mixed Lymphocyte Reaction
MRD	Minimal Residual Disease
N.A.	Not Applicable
ND	Not Done
NK	Natural Killer Cell
NO.	Number
NYHA	New York Heart Association
OMP	Orphan Medicinal Product
OS	Overall Survival
PB	Peripheral Blood
PBMC	Peripheral Blood Mononuclear Cell
PM	Project Manager
PSCT	Allogeneic Peripheral Stem Cell Transplantation
QA	Quality Assurance
QP	Qualified Person

SAE	Serious Adverse Events
SITC	Society for Immunotherapy of Cancer
SOC	Standard Of Care
SOP	Standard Operation Procedure
SQV	Site Qualification Visit
SUSAR	Suspected Unexpected Serious Adverse Reaction
TAA	Tumour Associated Antigen
TEAE	Treatment Emergent Adverse Event
TMF	Trial Master File
TNF	Tumour Necrosis Factor
TTR	Time To Remission
VU	Vrije Universiteit / Free University
WBC	White Blood Cells
WHO	World Health Organization
WT	Wilms Tumour Gene

6 SYNOPSIS

Name of Sponsor/Company: DCprime bv	
Name of Investigational Product: DCP-001	
Name of Active Ingredient: The active ingredient consists of allogeneic human dendritic cells (DCs) derived from the CD34+ progenitor AML cell line, DCOne. These DCs endogenously express several AML specific leukemia associated antigens (LAA) that include WT1, MUC1, PRAME and RHAMM.	
Title of Study: Allogeneic Dendritic cell Vaccination in AML oNgoing Clinical Evaluation II	
Short Study Title (acronym): ADVANCE II	
Study center(s): Approximately 3-5 sites Countries: Netherlands, Germany	
Coordinating Investigators: Prof. Dr. Arjan A van de Loosdrecht	
Studied period (years): Recruitment time: 12 - 18 months. Treatment time: 4.5 months Follow up time: 12 months after the last patient receives the last biweekly (4th) vaccination. Total Study duration: approximately 34 months	**Phase of development**: IIa, Proof of Concept
Rationale: Immunotherapy offers promise in AML and has created opportunities for improved outcomes. Dendritic cell (DC)-based immunotherapy has shown to be a promising strategy for the elimination of minimal residual disease (MRD) in patients with acute myeloid leukemia (AML). DCP-001 is an allogeneic dendritic cell based immunotherapeutic vaccine that expresses a number of tumour antigens that are expressed in AML and have shown clinical results when applied as peptide vaccines or loaded onto patient derived DC. Evidence for DCP-001 mediated safety, feasibility and immunological responses have been demonstrated in a Phase I study; the current study aims to generate evidence for clinical efficacy.	
Objectives: **Primary:** • To assess the effect of DCP-001 on MRD. MRD will be measured by flow cytometry pre and post vaccination as a surrogate marker for established clinical endpoints in AML. • To assess the effect of DCP-001 on immune responses in AML patients in first complete remission (CR1) and persistent MRD. • To document safety and tolerability.	

Secondary:

- To identify surrogate (immunological) markers that might correlate with clinical outcome.
- To quantify any lag time between initiation of treatment & onset of effect.
- To determine the effect of DCP-001 on Time to Relapse (TTR).
- To determine the effect of DCP-001 on Overall Survival (OS) during the study.
- To document the difference between the two vaccination regimens based on MRD outcome and immunological parameters.

Methodology:

International, multicentre, open-label proof of concept study without randomization and stratification. Two different dose groups are included. Group 1 consists of 10 patients that will receive 25E6 DCP-001 cells per vaccination with two additional booster vaccinations of 10E6 cells. Group 2 consists of 10 patients who will receive 50E6 DCP-001 cells per vaccination with two additional booster vaccinations of 10E6 cells.

Patients will be screened for eligibility for the study and evaluated at baseline, at each vaccination visit and every 8 weeks during follow up. Each patient will be followed up for 12 months after the 4th vaccination. Sera and cell samples (blood and bone marrow) will be collected when indicated for efficacy (MRD evaluation) and immune response monitoring.

Number of patients (planned):

No less than 20 patients are planned for participation

MRD assessment:

MRD will be determined at start of the vaccination, prior to the first boosting vaccination, after the last boosting and a final MRD assessment will be performed 100 days after receiving the last boost (day 224). MRD will be determined with multi-parametric flow cytometry. Flow cytometric MRD is based on leukaemia associated phenotype (LAP) that will be established at diagnosis and at specified time points post-vaccination. MRD will be reported as a percentage of the whole white blood cell compartment in complete remission, according to standardized protocols. Assessment of MRD will be performed by a central lab (VUmc).

Immunomonitoring:

- Antigen-specific T cell mediated immunity will be measured in PBMC by ELISpot and Intracellular cytokine release against antigens known to be expressed by DCP-001 (i.e. PRAME, WT-1, MUC1, RHAMM), both pre-and at various time points post-vaccination;
- PBMC profiling will be performed both pre-and at various time points post-vaccination;
- The overall immune competence of patient's PBMC to respond to DCP-001 will be measured both pre-and at various time points;
- Biopsies will be taken from the injection site for immunohistochemistry analyses 48 hrs after the 1st vaccination and 48 hrs after the 4th vaccination.

Diagnosis and main criteria for inclusion:

Main Inclusion Criteria:

1. Confirmed diagnosis of AML according to WHO2016 criteria, including cytological, molecular and cytogenetic criteria (except acute pro-myelocytic leukaemia/APL).

2. In CR1 (first complete remission) or CRi (incomplete blood count recovery) documented by bone marrow examination up to one month before vaccination; CR defined as less than 5% blasts in normo-cellular bone marrow, ANC >1*E9/L, platelet count >100*E9/L, no evidence of extra-medullary disease. Patients in CRi (patients with <5% blasts but with incomplete blood count recovery) should have platelets >50 E9/L.
3. MRD as defined by multicolour flow cytometry (MFC) at a value of > 0.1%, or detection of specific molecular abnormalities such as NPM1 mutation.
4. Patients that are in CR1 or CRi. Patients not having undergone consolidation therapy must have been in CR1 for at least 1 month prior to enrolment.
5. Expected and willing to undergo all study procedures, including outpatient evaluations for clinical and immunological monitoring.
6. Male or female of ≥ 18 years of age.
7. Women of childbearing potential must be using anti-conceptive therapy or use two (2) barrier contraceptive methods (one by each partner and at least one of the barrier methods must include spermicide (unless spermicide is not approved in the country or region). See section 12.8 for birth control methods deemed acceptable for this study.
8. ECOG (WHO) performance status 0-2.
9. Willing and able to provide written informed consent for participation in the study

Main Exclusion Criteria:

1. Acute Promyelocytic (APL; M3) type of AML.
2. Patients who have undergone or are scheduled for allogeneic stem cell transplantation.
3. History of previous allogeneic bone marrow or solid organ transplantation.
4. Uncontrolled or serious infections
5. Ongoing immunosuppressive therapy, other than short use of low dose steroids, i.e. equivalent to an average dose of ≤10mg of prednisone/day.
6. Chemotherapy and antineoplastic hormonal therapy within 28 days prior to the screening visits.
7. Active autoimmune disease.
8. Inadequate liver function (AST and ALT > 3 x ULN, serum bilirubin >3 x ULN).
9. Other active Malignancies within the last 5 years, except for adequately treated carcinoma in situ of the cervix or squamous carcinoma of the skin or adequately controlled limited basal cell skin cancer.
10. Pregnant or lactating females.
11. Major surgical procedure (including open biopsy) within 28 days prior to the first study treatment, or anticipation of the need for major surgery during the course of the study treatment.
12. Uncontrolled hypertension (systolic > 150 mm Hg and/or diastolic > 100 mm Hg) or clinically significant (i.e. active) cardiovascular disease.
13. Evidence of any other medical conditions (such as psychiatric illness, physical examination or

laboratory findings) that may interfere with the planned treatment, affect patient compliance or place the patient at high risk from treatment-related complications.

14. Known HIV, Hepatitis B and/or Hepatitis C infections.
15. History of hypersensitivity to the investigational medicinal product or to any excipient present in the pharmaceutical form of the investigational medicinal product.

Investigational product, dosage and mode of administration:

The investigational product is DCP-001, which consists of allogeneic human dendritic cells, expressing specific Leukaemia Associated Antigens (LAA) antigens.

Patients will receive either 25E6 (Group 1) or 50E6 (Group 2) DCP-001 cells/vaccination. Both groups will receive a total of 4 vaccinations at biweekly intervals (weeks 0, 2, 4 and 6) followed by 2 booster vaccinations 8 and 12 weeks after the 4th (week 6) vaccination of 10E6 cells. DCP-001 will be applied intra-dermally (2-4 injections per vaccination, 1 per booster vaccination) and is formulated as a direct injectable vaccine.

Duration of treatment:

4 vaccinations at biweekly intervals, followed by two booster vaccinations 8 and 12 weeks after the 4th (week 6) vaccination. Total treatment duration 18 weeks.

Reference therapy, dosage and mode of administration:

N.A.

Criteria for evaluation:

1. Any change in MRD (flow cytometric) as compared to baseline MRD
2. Any change in immunoreactivity (specific and non-specific) as compared to baseline.

Safety endpoints:

1. Treatment emergent adverse events (TEAEs) will be collected in the first 48 hours after the first vaccination to determine (acute) toxicity due to intradermal injection.
2. All TEAEs will be collected during the study for each patient and the grade of toxicity using Common Terminology Criteria for Adverse Events (CTCAE V4) and relationship to product determined.
3. A Data Safety Monitoring Board (DSMB) will meet after every 5 patients treated to evaluate safety; written reports will be made of each meeting.
4. All Serious Adverse Events (SAEs) will be reported by the investigator within 24 hours of first knowledge. All SAEs will be reported in a timely manner to the appropriate regulatory body, consistent with existing regulations. Information on relevant SAEs will be disseminated between sites in a timely manner.
5. The safety and efficacy databases will include only the subjects included in study and received a least one vaccination.

Statistical Methods:

Sample Size Assumptions: The planned sample size of 20 evaluable subjects will be distributed 1:1 with 10 subjects each in Group 1 - 25E6 DCP-001 cells per vaccination, and Group 2 - 50E6 DCP-001 cells per vaccination. Additional subjects may be enrolled to ensure that 20 evaluable subjects complete the study. The sample size is based on practical considerations, rather than strict sample size statistical considerations with a desired power for a pre-specified difference.

Statistical Analyses: Descriptive analyses of efficacy and safety variables will be used. The analysis will be performed on the data collected up to the last MRD assessment (Day 224). In general, descriptive statistics, unless otherwise noted, will include the number of subjects, mean, standard deviation, median, minimum value, and maximum value, and where applicable, 95% confidence intervals. Percentages will be calculated using the number of subjects within each treatment group. If appropriate, immune monitoring data will be summarized using the geometric mean, standard error of the geometric mean, and 95% confidence intervals of the geometric mean. Time to event variables (e.g. time to relapse or change in MRD status) will be analysed using survival analysis techniques (e.g, Kaplan-Meier estimates).

7 FLOW CHART AND SCHEDULE OF ASSESSMENTS

Table 1: Flowchart and assessment schedule

	Screening	Baseline wk 0	wk 0	wk 2	wk 4	wk 6	wk 6	wk 11	wk 14	wk 18	wk 18	wk 20	wk 32	FU[1] +8 weeks	End of Study[2] (Final FU Visit)
days	-28	0	2[7]	14[8]	28[8]	42[8]	44[7]	77[8]	98[8]	126[8]	128 [7]	140[8]	224[8]		
Visit No.	1	2	3	4	5	6	7	8	9	10	11	12	13	14-17	18
Informed consent	√														
Inclusion/exclusion criteria	√														
Demographics	√														
Medical History	√														
Physical examination	√	√		√	√	√		√	√	√		√	√	√	√
ECOG	√	√		√	√	√		√	√	√		√	√	√	√
HLA Typing[11]	√														
Vital signs	√	√[3]		√	√	√		√	√	√		√	√	√	√
ECG, X-thorax	√														
Haematology[4]	√	√[3]		√	√	√			√	√		√	√	√	√
Biochemistry[5, 13]	√	√[3, 13]		√	√	√			√	√		√	√	√	√
Urine analysis[10]	√														√
Viral tests (HIV, Hepatitis B and C)[12]	√														
MRD assessment (BM aspirate)		√							√			√	√		
DCP-001 Vaccination		√		√	√	√			√	√					
Immunomonitoring		√	√				√	√			√	√	√		
Skin biopsy[9]			√				√				√				
End of treatment													√		
End of study															√
Prior and Concomitant medication	√	√	√	√	√	√	√	√	√	√	√	√	√	√	√
Adverse event reporting[6]		√	√	√	√	√	√	√	√	√	√	√	√	√	√

1) Follow up visits on wk 40, 48 and 56
2) End of study visit (last FU visit) will be performed 12 months after 4th vaccination, or, no more than 2 weeks after last treatment received if a patient goes off study before week 32
3) If baseline assessment is within 7 days of first vaccination and if results meet the stated eligibility criteria, these assessments need not to be repeated on day 0
4) Hemoglobin, Ht, erythrocytes, reticulocytes, platelets, WBC and differential
5) Na, K, creatinine, Ca, P, Mg, bilirubin, AF, GGT, AST, ALT, LDH, glucose, APTT, INR, fibrinogen
6) Until 30 days after last vaccination/booster and thereafter as long as DCP-001 related adverse events are ongoing
7) Days are fixed with the previous visit
8) Days ± 3 days
9) Biopsies will be taken from the injection site for immunohistochemistry analysis 48 ± 6 hrs after the 1st vaccination and 48 ± 6 hrs after the 4th vaccination
10) pH, Protein, erythrocytes, leucocytes, glucose, ketones, nitrites
11) HLA-A, HLA-B, HLA-C, HLA-DR, HLA-DQ
12) HIV, Hep B and Hep C serology tests (Anti-HIV 1+2, HBsAg, Anti HBc-IgG, anti HBc-IgM and Anti-HCV)
13) Beta hCG blood test in woman with child-baring potential

8 INTRODUCTION

8.1 SUMMARY

Acute Myeloid Leukaemia (AML) is a clonal disorder arising from hematopoietic progenitors developing in the myeloid lineage, characterized by deregulated differentiation and maturation programs. AML most commonly affects the elderly population with a median age of around 67 years (1,2). Current AML treatment relies largely on intensive chemotherapy and allogeneic hematopoietic stem cell transplantation (HSCT), which unfortunately is unsuccessful in 60-80% of patients due to persistence of minimal residual disease (MRD) (3,4). Particularly in elderly patients, these strategies are associated with high co-morbidity rates, and the 5-year overall survival in this population remains poor (10-15% in patients > 65 years of age). For this reason, new treatment strategies are urgently needed.

A highly promising approach to meet this challenge is to develop immunotherapeutic interventions that stimulate the immune system to recognize remaining leukemic (stem) cells as foreign, and eliminate them. Indeed, immunotherapy through active vaccination is expected to have great potential in eradicating MRD, and the purpose of this study is to investigate an emerging form of immunotherapy for post-remission AML patients in a Phase IIa clinical study. The therapy consists of vaccination with the dendritic cell based DCP-001 vaccine formulation, and is based on a successfully completed Phase I feasibility and safety study (see section 8.7.1). DCP-001 is crucially different from earlier DC-based therapies in AML, in that it combines the antigen carrier properties of a leukaemia cell line with the immune-stimulatory properties of DC. It is derived from an AML cell line and endogenously expresses multiple leukaemia-associated antigens, inducing a multi-targeted immune response. It can be applied as an off-the-shelf allogeneic product and is irradiated and frozen for direct use. This DC-leukaemia cell vaccine formulation, DCP-001, is designated as an Orphan Medicinal Product (OMP) in the European Union (EU/3/12/969) for treatment of AML patients post-remission.

8.2 DESCRIPTION OF THE CONDITION

Acute Myeloid Leukaemia (AML) is a disorder arising from the myeloid lineage of white blood cells, leading to accumulation of immature blasts cells in the bone marrow and peripheral blood, and disruption of normal haematopoiesis. The disease is heterogeneous with respect to morphology, immune-phenotype, cytogenetic and molecular genetics, clinical characteristics and outcome.

AML is relatively rare, afflicting annually 3–5 cases per 100,000 persons but remains the most common form of acute leukaemia among adults. As also the deadliest form, it accounts for the largest number of annual deaths in the United States and Europe. With a median age at diagnosis of 67 years, this disease is far more common in the elderly (1). In this age group, AML has a particularly dismal outcome with less than 10% of the patients being alive 5 years after the diagnosis, as compared to 40% in the young (1,5). With an age dependent rise in incidence and a peak at ~22 cases per 100,000 in the geriatric population, AML has a devastating impact on the survival of this age group.

The conventional initial treatment of AML is intensive chemotherapy followed by an autologous/allogeneic hematopoietic stem cell transplantation (HSCT). Due to co-morbidity in the elderly patients, some cannot tolerate aggressive chemotherapy and, in patients ineligible for induction chemotherapy, current treatment options are focused mainly on supportive measures, such as red blood and platelet transfusions when needed and antibiotics for infections. Even when induction chemotherapy is able to induce complete remission, relapse rates remain high (60-80%) due to the

presence and outgrowth of residual AML cells in bone marrow (BM); so called minimal residual disease (MRD), which directly relates to the poor survival rates.

The need for a suitable treatment to eradicate MRD and improve disease outcome is therefore an important focus area for new therapeutic approaches in AML, with no other means available at the present time. Active immunotherapies in particular are being explored as treatment options for haematological tumours, and these aim to induce endogenous tumour immune surveillance mechanisms by exposing the patients' immune system to tumour-associated antigens (TAAs) delivered in an immunogenic form. Such vaccines act by eradicating MRD after activation of CD8+ cytotoxic T-lymphocytes (CTL), and anti-leukemia CTL are most effectively primed by the most powerful antigen-presenting cell identified to date, i.e. the Dendritic Cell (DC). For this reason, many studies have explored the efficacy of DC-based cancer therapies in both AML and other malignancies, and these will be reviewed below.

8.3 DENDRITIC CELL PRODUCTS AS THERAPEUTIC CANCER VACCINES

DCs are specialized to capture and present antigens (TAAs, and all other antigens, for that matter), converting the proteins to peptides that are presented through MHC class I and II pathways and initiate antigen specific T-cells responses (6). These antigen specific T-cells in turn can attack tumour cells expressing the antigenic determinants, or can contribute to stimulation of B-cell responses which produce antibodies and, in some cases, lead to tumour cell death (7).

Multiple clinical trials have shown that DC vaccination with patient-derived dendritic cell vaccines endowed with TAAs of choice, is safe, has hardly any side effects, and is associated with the induction of immune responses (8,9). Nevertheless, although proof of concept for DC-based therapy has been provided in the autologous DC-setting, this autologous approach is cumbersome, because of the logistic complexity of harvesting and processing the patients' own cells. In addition, it is costly, has quality issues and is difficult to scale up, which precludes broad application. This forms the basis for the development of the dendritic cell product DCP-001 used in the present clinical protocol, which is a DC-based vaccine that lends itself to be developed as an off-the-shelf product which expresses multiple TAAs (see below).

As for mode of action: whereas initial research with DC-based vaccines focused on their ability to generate CD4 and CD8 T cells responses specific for TAAs, resulting in tumour specific T cells responses, it has become increasingly clear that the activation of multiple other immune effector cells is the key to success for curative cancer vaccination. This requires a coordinated induction of innate and adaptive immune responses including NK-cells, B-cells, CD4 and CD8 T-cells responses. DCs have the ability to induce such multi-functional immune responses by the induction of multiple pleiotropic cytokines and chemokines, all involved in recruitment and polarization of T-cell responses and activation of cell types like NK cells and myeloid cells.

It has also become clear that immune system priming may occur in 2 different manners with DC vaccines. First, injected DCs migrate from the site of injection to the lymph nodes, where they trigger T-cell responses. Such a process is referred to as direct priming. Secondly, it is known that only low numbers of injected DC actually reach the lymph nodes, varying between 1-5% of the injected DCs (10-12). It is therefore presumed that the highly effective induction of T cell responses upon DC vaccination is also due to the participation of host dendritic cells, which can take up antigens from the injected DC at the injection site, and through that mechanism present antigens in a process referred to as indirect or cross priming (13).

8.4 RATIONALE FOR USING THE AML-DERIVED DC LINE DCONE AND MODE OF ACTION

The DC-based product used in the present clinical protocol is called DCP-001, and represents the first next-generation DC vaccine. It lends itself uniquely to off-the-shelf application, and combines the immune-stimulatory features of allogeneic DC vaccines and multi-antigen vaccines. Allogeneic DCs have been reported to induce stronger antigen-specific immune responses than autologous DCs (14,15), because they trigger a broader T-cell immune repertoire, plus a broad inflammatory response. This precludes the requirement for adjuvant.

The DCP-001 vaccine formulation that will be tested in this study, has been tested in an open label, Phase I, feasibility and safety, dose escalation study (NCT01373515), without stratification or randomization, conducted in 12 elderly AML patients not eligible for additional intensification or SoC therapies. AML patients were enrolled who were either in complete remission (CR1/CR2) (n=5) or had smouldering disease (n=7). The primary objectives of the study (feasibility and safety) were achieved, with 10/12 patients completing the vaccination program. Treatment was well tolerated, and only one serious adverse events related to DCP-001 was observed. A clear-cut distinction was noted between patients with and without detectable circulating leukemic blasts during the vaccination period. Patients with circulating blasts died within 6 months post-treatment, patients without showed unusually prolonged survival (median overall survival 36 months (range 7-65) from start of vaccination treatment. Long- term survival was correlated with maintained T-cell levels and induction of multi-functional immune responses. Thus, vaccination with DCP-001 is safe and feasible in AML patients, and generates both cellular and humoral immune responses. Together with the above described characteristics of its mode of action, these clinical results from the rationale behind the intended Phase IIa.

8.5 DESCRIPTION OF THE INVESTIGATIONAL PRODUCT

The active substance in DCP-001 consists of allogeneic human dendritic cells derived from the AML cell line DCOne. DCP-001 functions as a combination of a dendritic cells vaccine and a whole tumour cell vaccine, endogenously expresses multiple undefined and defined leukaemia antigens, including WT-1, PRAME, RHAMM, and MUC1, all proven tumour targets in both AML as well as other tumours (6). Due to the endogenous expression of these antigens, together with the expression of costimulatory molecules, essential for effective T cell activation, DCP-001 has the capability to stimulate broad T-cell responses against all of these antigens that also exhibit the ability to kill tumour cells. It is known that multi-targeted immunotherapy is crucial for success.

8.6 NON - CLINICAL PHARMACOLOGY/PHARMOKINETICS AND TOXICOLOGY

DCP-001 is a sterile cell suspension for intra-dermal injection consisting of allogeneic human dendritic cells (DCs) derived from a CD34+ progenitor cell line (DCOne). DCOne is a well-characterized cell line from human origin. The DCs endogenously express several AML specific leukemia associated antigens that include WT1, MUC1, PRAME and RHAMM. DCP-001 is manufactured by expanding and maturing the DCs from DCOne without further (genetical) manipulations.

The non-clinical development of DCP-001 was designed on a risk-based strategy with the aim to demonstrate proof-of-principle and to substantiate human response to an extent where it is deemed appropriate to move forward into clinical development. A Phase I, feasibility and safety dose escalation study in 12 AML patients has already successfully been completed.

Pharmacology studies / Mode of action

It is acknowledged by regulatory bodies that the absence of relevant non-clinical models with good predictive properties constitutes a great hurdle for efficient drug development of anticancer medicinal products (EMA guideline EMA/CHMP/205/95/Rev4). A wide range of in vitro potency and phenotypic/functional characterization assays have been applied substantiating the mode of action of DCP-001 and showing that DCP-001 holds the main characteristics of DC cells to exert their immunogenic effect (see Investigator Brochure; (EMA/CHMP/BWP/271475/2006 rev. 1).

Data from the Phase I clinical study confirm that DCP-001 is immuno-active as indicated by clinical evidence for multifunctional and lasting immune responses.

Pharmacokinetics and biodistribution

No specific pharmacokinetic, metabolism or bio distribution studies in animals have been performed on DCP-001. It has been confirmed in regulatory body meetings with EMA and FDA that these studies are not warranted for these types of products. It is in this respect worthwhile to mention that DCP-001 cell suspension is injected intra-dermally. This routing cannot be mimicked in mice. In addition, it is emphasized that the effect of DCP-001 is intended to originate solely from the matured DCs in DCP- 001 and not from in vivo proliferation in the host after administration. Gamma irradiation of cells during manufacturing of DCP-001 has been applied to effectively arrest any proliferative cellular capacity while retaining viability and biological activity.

Toxicology / Safety

The difficulty to use a conventional nonclinical development approach in order to predict human response and especially safety is recognized for anticancer medicinal products and also for tumour vaccines and cell-based therapies (EMEA/CHMP/205/95/Rev.4; EMEA/CHMP/410869/2006; ICH S9 EMEA/CHMP/ICH/646107/2008). The injection of a human cell based product in any type of experimental animal will lead to a fast-immune attack eradicating the human cells in a few days. In addition, while nonclinical models have provided the basis for understanding the mechanisms of immunotherapy, immunological reactions are difficult to predict and translate from animals to humans (Society for Immunotherapy of Cancer (SITC) with members form Europe, Japan, China and North America (16).

From the above, it is clear that for human cellular products such as DCP-001, the conventional toxicology program with single dose, repeated dose, local tolerance testing etc., is not applicable. This lack of relevant pharmacokinetic and safety data in animals, is countered by the results of a Phase I study with DCP-001 in which no drug-related safety or toxicity issues have been observed. This mirrors the current extensive clinical experience with multiple products with a similar composition, such as allogeneic GVAX vaccines, autologous DC based vaccines produced from patients' DC, allogeneic DC from healthy donors, and allogeneic whole cancer cellular vaccine. All of these have documented solidly that no safety issues occur.

8.7 CLINICAL DATA

The first human study of DCP-001 has been successfully finalized. This study enrolled 12 AML patients in a 3+3 dose escalation design with extension to 6 patients in the highest non-toxic dose level. Main criteria for inclusion were either AML in 2nd complete remission (CR2), (relapsed) smouldering AML, or de novo AML in CR1, all not eligible for intensification therapy due to co-morbidity and poor performance status.

8.7.1 Study DCOne-001

EudraCt number: 2008-006950-16

Methodology

This was a single centre, open label, Phase I, feasibility and safety, dose escalation study, without stratification or randomisation, conducted in 12 patients chosen according to the following criteria:

- Patients with AML in second CR, not eligible for additional intensification therapies; or
- Patients with relapse (smouldering) AML not eligible for additional intensification therapies; or
- Patients with de novo (smouldering) AML not eligible for intensive treatment according to current HOVON trials; or
- Patients >65 years of age with de novo AML in first CR and off protocol of current HOVON protocols.

Main exclusion criteria included uncontrolled active infection, previous immunotherapy in the last 3 months, and previous allogeneic peripheral stem cell transplantation (allo-PSCT). Patients received 4 vaccinations during the study, administered in a bi-weekly schedule. The first cohort (n=3) received 10E6 DCP-001 cells/vaccination, the second cohort (n=3) received 25E6 DCP-001 cells/vaccination, and the third cohort (n=3) received 50E6 DCP-001 cells/ vaccination. This cohort was extended with additional 3 patients after confirmation that this dose was not toxic.

Patients were treated in the outpatient clinic and vaccinated with DCP-001 at days 0, 14, 28 and 42. Skin testing was performed at days 2, 49 and 51. A final visit took place at the end of study at day 126. During the treatment phase, (S)AEs and concomitant treatments were recorded continuously.

Extensive immune monitoring at baseline (day 0), one week after last vaccination (day 49) and at follow-up (day 126) was performed to assess immunogenicity of the DCP-001 vaccine and ascertain any relationships to clinical outcome.

Safety and feasibility

The safety, feasibility, and biological effects of DCP-001 vaccination were investigated in 12 elderly AML patients. Patient characteristics are listed in Table 2). Twelve patients, 3 per cohort, were enrolled and treated. Patients (age range 57-74) were either in CR1/CR2 (n=5) or had smouldering disease (n=7). Patients had received all available standard care, were at high risk of relapse and not eligible for available post-remission therapies, including allogeneic stem cell transplantation. Twelve patients initiated vaccination of which 10 patients received all 4 vaccinations and two patients (patients 002 and 011) received 3 vaccinations. One patient discontinued due to disease progression (pat 002) and one patient (pat 011) due to a candida endocarditis.

DCP-001 vaccination was well tolerated, safe and feasible with 10 of 12 patients (83.3%) completing the vaccination program. Six patients experienced serious adverse events (SAE) during the study (patients 002, 004, 005, 007, 011 and 014), only one was judged to have a possible relationship to study treatment (patient 002 with diabetes insipidus). This could possibly represent a vaccine induced autoimmune response. All others were considered unrelated or unlikely to be related to study treatment. The diabetes insipidus occurred 4 weeks after the 3rd vaccination and after treatment had already been discontinued due to disease progression.

Two patients died before completing the study due to pneumonia (pat 005) and disease progression (pat 014). Neither of these deaths were reported as related to study treatment. Two patients discontinued

study treatment (see above); patient 011 due to the SAE (endocarditis) and patient 002 due to disease progression (Note: pat 002 also had an SAE [diabetes insipidus] after having discontinued treatment). SAEs in patients 004 (myocardial infarct) and 007 (vasovagal collapse) did not lead to discontinuation and both patients in fact proved to be long-term survivors (see Figure 1). The distribution of AEs between cohorts was uneven but there was no trend suggesting a dose-response relationship for AE occurrence. AEs were of CTC-grade 1 (mild) or CTC-grade 2 (moderate) and most were judged as unrelated to study treatment (70 of 98 AEs). The most common AEs were injection site reactions (6 patients), anaemia (4 patients), thrombocytopenia (3 patients), fatigue (3 patients), pain in extremity (3 patients), and nausea (3 patients). The most common preferred terms that were judged as related (possibly, probably or definitely) to IMP were injection site reaction (4 patients), injection site erythema (3 patients), pruritus (2 patients), pain in extremity (2 patients).

Local effects at the site of injection were reported by 7 of 12 patients (10 events in total). This included: injection site reaction (5 patients), injection site erythema (3 patients), and injection site induration (1 patient). The local effects were reported by patients in Cohorts 1, 2 and 4. All events were judged to be definitely related to IMP and to be related to the first vaccination except for one event which was judged to be definitely related to the second vaccination and one event of injection site reaction (vasovagal collapse) which was judged as unrelated to IMP. All local effects (except for the vasovagal collapse) were of CTC-grade 1 (mild) and resolved completely within 3 to 26 days. In general, the local effects appeared 2 days after the injection. Changes in laboratory parameters occurred in individual patients. There were no trends suggesting a dose-response relationship with IMP.

No safety concerns of DCP-001 vaccination were identified based on the AE reporting, laboratory assessments, physical examinations, vital signs or weight assessments in the study.

Table 2: Patient characteristics

Patient nr	Age	Sex	Time between AML diagnosis and study entry (Mo)	Disease status		Dead/alive at end of study	% Blasts in Bone Marrow (cytology)	
				at study entry	at end of study		at study entry	at end of study
DC-001	66	F	8	AML relapse/Smouldering	CR	Alive	7	3
DC-002	70	M	7	AML relapse/Smouldering	Disease progression	Dead	53	89
DC-004	72	F	45	AML in CR2	CR	Alive	2	3
DC-005	74	M	75	AML relapse/Smouldering	Pneumonia	Dead	58	M[1]
DC-006	69	F	18	AML relapse/Smouldering	Disease progression	Alive	14	80
DC-007	74	F	14	AML relapse/Smouldering	Smouldering disease	Alive	M	7
DC-008	64	F	7	de novo AML, in CR1	CR	Alive	0	M
DC-011	57	M	11	de novo AML, in CR1	endocarditis	Dead	1	M[1]
DC-012	70	M	22	AML relapse/Smouldering	Disease progression	Alive	5	31
DC-013	65	M	3	de novo AML, in CR1	CR	Alive	2	2
DC-014	67	M	20	AML relapse/Smouldering	Disease progression	Dead	29	M[1]
DC-015	68	M	5	de novo AML, in CR1	CR	Alive	7	1

Clinical outcome

At the end of study (day 126), 9 out of 12 patients were alive. Eight patients completed all assessments and for those 4 patients who did not complete the study, reasons were disease progression (patient 002), death due to disease progression (patient 014), pneumonitis / pneumonia (patient 005) and candida endocarditis (patient 011).

Six out of 12 patients were in complete remission (i.e. undetectable AML blasts in blood and < 5% in bone marrow) at end of study and 5/12 experienced persistent disease (Table 2). All patients, except for one, who were in CR at end of study were in CR1 or CR2 at baseline. All *but one* of the patients with smouldering disease at study entry, had persistent disease at the end of the study. One patient (pat 001) with smouldering disease reached CR at the end of the study. This patient had circulating blast counts of $0.05x10^9$/L and bone marrow blasts of 7% at baseline. No clear relationship between clinical outcome and administered DCP-001 dose was apparent.

Follow up

All patients were followed until death. Based on the presence or absence of circulating blast cells patients could be divided into 2 groups, which turned out to correspond to short- (≤6 months, median overall survival 3 months, range 2-6 months) and long-term survivors (>6 months, median overall survival 36 months, range 7-63 months), respectively (Figure 1). The two groups showed strikingly different patterns in peripheral leukemic blast and T-cell counts during treatment (Figure 2). Patients with detectable and persistence of peripheral blast cells died within 6 months post-treatment (short- term survivors). Patients without detectable leukemic blasts in peripheral blood (or rapidly dropping below the detection threshold) showed remarkably prolonged survival with one patient still alive at the time of writing and 54 months after study entry and the other patients surviving for 7, 36, 22, 63, and 36 months (Figure 1). Long-term survival was accompanied by maintained levels of T-cells. In patients with short-term survival and detectable circulating blast cells, T-cell levels dropped precipitously (Figure 2).

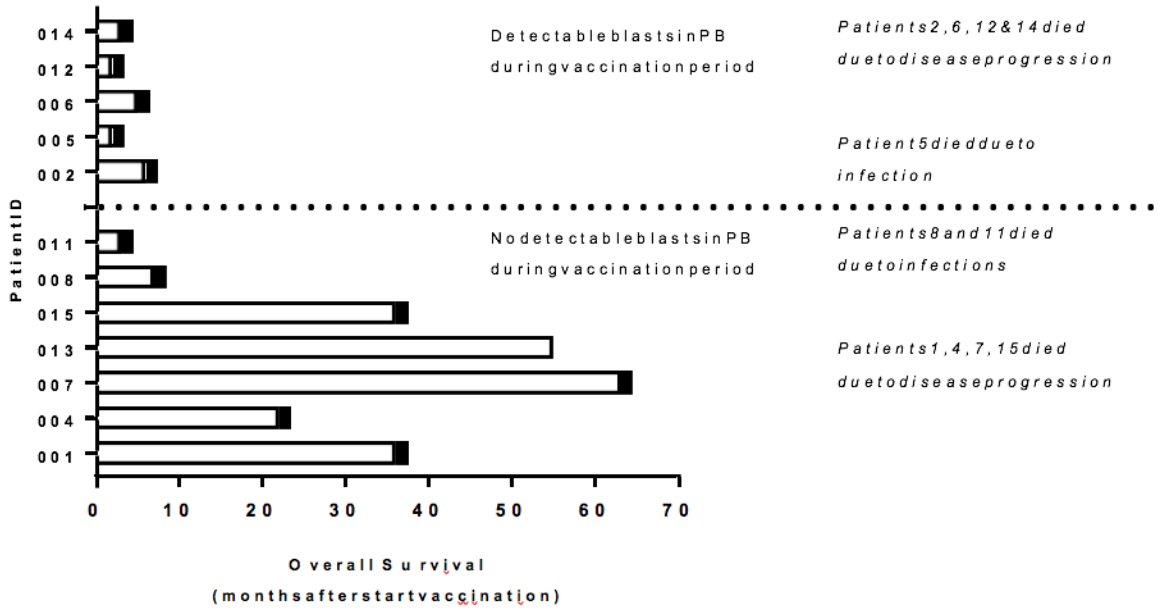

Figure 1: Detectability of leukemic blasts in patients is related to post-vaccination survival. Shown is overall survival since start of vaccination; patients were subdivided by the presence of detectable leukemic blasts in peripheral blood during treatment (dotted line). Death is indicated by black box; causes of death are listed.

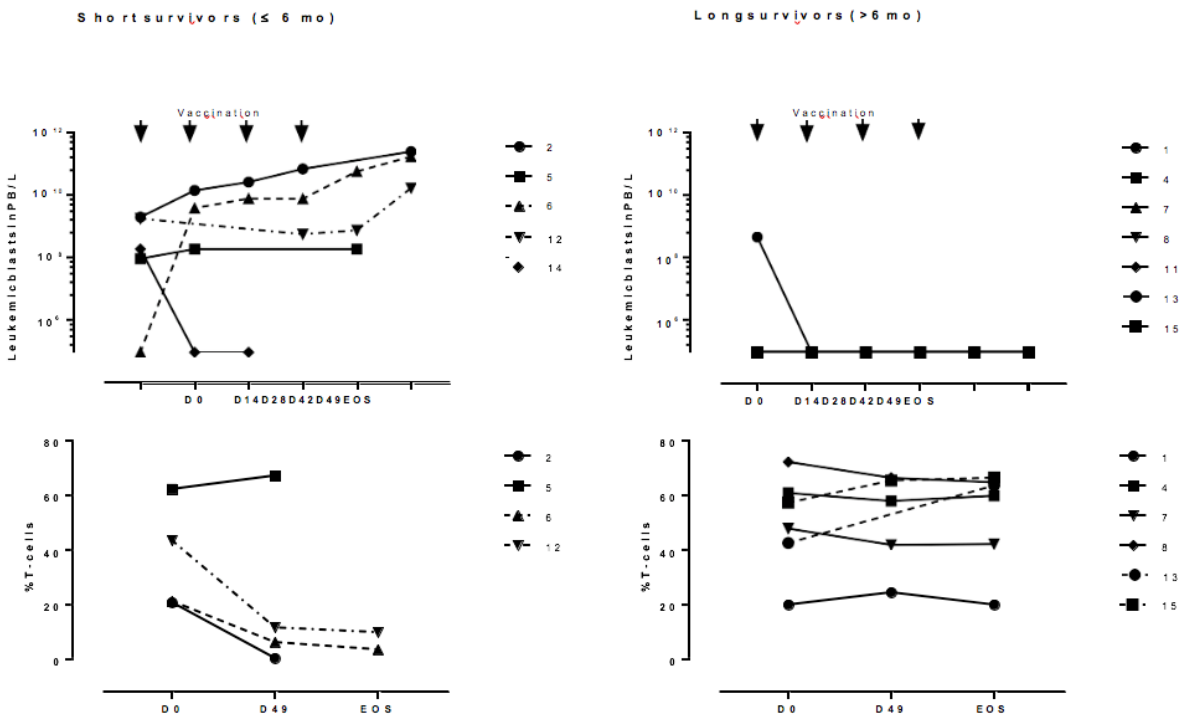

Figure 2: Leukemic blast and T cell rates in short- versus long-term survivors. Absolute numbers of leukemic blasts in peripheral blood (PB) over treatment (in days, EOS=end of study) in A) short-term (less than 6 months) survivors and in B) long-term (more than 6 months) survivors. T cell rates (as percentage of PB mononuclear cells) in C) short-term (less than 6 months) survivors and in D) long-term (more than 6 months) survivors.

Immune response monitoring

Extensive immune monitoring at baseline (day 0), one week after last vaccination (day 49) and at follow-up (day 126) was performed to assess immunogenicity of the DCP-001 vaccine and ascertain any relationships to clinical outcome. Patients with detectable leukemic blasts at study entry (i.e. patients 002, 005, 006, 012, and 014) experienced rapidly dropping T-cell frequencies (Figure 2) which in some instances precluded *in-vitro* T-cell-based monitoring at days 49 and 126. An overview of functional T-cell-related monitoring data is provided in Table 3.

Table 3: Overview T-cell immune monitoring data

	Patient ID*	Δ T-cell response to DCP-001	TAA-specific T-cell response	Δ T-cell influx at DTH site	Δ DTH	Overall T-cell Score***	Survival Post-Vacc (months)
Short survivors ≤6 months	002	ND	-	-	-	0/3	6
	005	-	ND	ND	+	1/2	2
	006	-	ND	+	-	1/3	5
	012	+	+	+	-	3/4	2
	014	ND	-	-	-	0/3	3
Overall responses		**1/3**	**1/3**	2/4	1/5	**5/15 (33%)**	**3.6**
Long survivors >6 months	001	-/+**	-/-**	+	+	3/4	36
	004	-	+	-	+	2/4	22
	007	-	-	+	-	1/3	63
	008	+	ND	+	+	3/3	7
	013	+	+	+	+	4/4	55

	015	+	+	ND	-	2/3	36
Overall responses		**4/6**	**3/5**	4/5	4/6	**15/22 (68%)**	**36.5**

* Patient 11 (3 months OS) was excluded from this analysis; died from infection, no monitoring data available
**Second data derived after two additional booster vaccines; T-cell response to DCP-001 was negative after first round of vaccinations
***Based on positive T-cell responses from evaluable data sets, *p*=0.049 by 2-sided Fisher's Exact test between ≤6 and >6-month survival groups
ND: not done

Antigen specific immune responses

T-cell reactivity by IFNγ ELIspot analysis after *in-vitro* stimulation was assessed against the AML-related antigens WT-1 and PRAME, both of which were expressed by the DCP-001 vaccine, as well as against NY-ESO-1 and MAGE-A3. NY-ESO-1 and MAGE-A3 were not expressed by DCP-001 but were included to monitor for possible epitope spreading. All tested patients showed pre-existent reactivity to the CEFT recall antigen pool, which was maintained over treatment, indicating good immune competence. Four out of eight evaluable patients showed DCP-001 induced or enhanced WT-1, PRAME, MAGE-A3, or NY-ESO-1 responses (Table 3).

Antibody responses

Antibody responses generated against DCOne progenitor and autologous blast antigens was evaluated in 10/12 patients. 5 patients showed an increased response denoting antigen-specific humoral responses. Induced antibody responses against autologous blasts were observed in two out of three evaluable patients, which demonstrates the induction of immunity against autologous AML blasts by the allogeneic DCP-001 vaccine.

DTH reactivity

In the DTH tests, 5 of 11 evaluable patients had an increase of ≥ 50% in the mean diameter of induration at day 51 compared to baseline (i.e. positive according to criteria formulated), indicating the induction of a T-cell-mediated response to DCP-001 (Table 3). No reactivity to the Cryostor vehicle control was ever observed (data not shown). Of the 7 patients who did not have an increase at day 51 compared to baseline, all showed a baseline reaction of at least 4mm (range 3.5-15mm). A measurable DTH response at baseline indicates pre-existing immunity to components of the DCP-001 vaccine. This could be an allo-reaction, but could also be a response to TAAs in the vaccine. Evidence for vaccination- induced increases in DTH reactivity was mostly found among long-term (>6 months) survivors (Table 3).

Immunohistochemistry

Immunohistochemical assessment of DTH punch biopsies was performed for T-cell and DC influx in 9 out of 12 patients at day 2 and day 51. All of these patients showed an increased influx of activated T- cells in response to the vaccine as compared to the Cryostor vehicle control.

T-cell reactivity to DCP-001 and its DCOne progenitors by MLR

T-cells of 9 out of 12 patients were analysed for their proliferative capacity in response to DCP-001 and DCOne progenitor cells. Seven out of 9 tested patients showed pre-existent T-cell responses (i.e. proliferation rate exceeding 20% of T-cells), as is to be expected for an allogeneic vaccine. Increased proliferative responses (either CD4 or CD8) against DCP-001 cells and/or DCOne progenitor cells upon vaccination were observed in 6/9 evaluable patients. Notably, in 5 cases increased post-vaccination reactivity to DCP-001 progenitors was observed, which almost exclusively involved $CD8^+$ T-cells, suggesting the induction of a $CD8^+$ effector T-cell response to AML blasts.

DCP-001-induced T-cell reactivity in relation to survival

Overall, the immune monitoring data clearly demonstrate DCP-001 induced T-cell reactivity. When results from the four functional T-cell related assays were combined, an "Overall T-cell Score" was derived (Table 3), which demonstrated significantly lower scores (5 positive assays out of 15 conducted) in short-term survivors (≤6 months)

versus long-term survivors. (>6 months, 15 positive assays out of 22 conducted, *p*=0.049 by 2-sided Fisher's Exact Test).

Conclusion

The primary objectives of the study (feasibility and safety) were achieved with 10/12 patients completing the vaccination program. Treatment was well tolerated. A clear-cut distinction was noted between patients with and without detectable circulating leukemic blasts during the vaccination period. Patients with circulating blasts died within 6 months post-treatment, patients without showed unusually prolonged survival (median overall survival 36 months (range 7-63) from start of vaccination treatment). Long-term survival was correlated with maintained T-cell levels and induction of multi-functional immune responses.

8.8 RATIONALE FOR STUDY

Allogeneic DC-based vaccination is a promising therapy option for the treatment of minimal residual disease in AML based on its mode of action as described above and the supportive results of the Phase I trial.

The next step in the development program is to try to obtain a clear efficacy signal based on a clinical parameter and to determine the magnitude of the clinical effect.

The primary endpoint for this study is to assess the effect of DCP-001 vaccination on MRD measured by flow cytometry as a surrogate marker for established clinical endpoints in AML pre- and post- vaccination. MRD evaluation is becoming the new standard in evaluating response in AML (17). Technical recommendations for MRD detection and its clinical use is supported by the European Leukemia Net (18).

The information obtained from this Phase II study will facilitate the design of a confirmatory Phase IIb study that includes linking of MRD to established clinical endpoints.

8.9 SELECTION OF DOSES IN THE STUDY

The Phase I study employed the traditional 3+3 design, starting with 10E6 cells per vaccination (3 patients), followed by 25E6 cells per vaccination (3 patients) and then 50E6 cells per vaccination (6 patients). In all three cohorts, immune responses were observed, as demonstrated by DTH responses, immunohistochemistry of biopsies of the DTH site, MLR, ELISpot and antibody responses. Not all parameters were measurable in all patients for a variety of reasons (e.g. insufficient material or technical difficulties). Preliminary analysis of the data indicated that the strongest immune responses were obtained in the middle cohort and highest dose cohort, but the study was too small to obtain a clear optimal dose signal between the mid and high dose.

Therefore, it the current study the safety and efficacy of both the mid (25E6 cells) and high dose (50E6 cells) will be established.

8.10 RISK-BENEFIT ASSESSMENT

In the Phase I study with DCP-001 the treatment was well tolerated, with no emerging potential safety issues

8.10.1 RISK OF INFECTION

Risk is considered low/absent as the DCOne cell line from which DCP-001 derived has been extensively tested for multiple viruses according to applicable regulatory guidelines (see IB). Upon release the DCP-001 product is confirmed to be sterile.

8.10.2 IMMUNE RESPONSE TO THE VACCINE

See section 8.7.1

8.10.3 INTRADERMAL INJECTION AND SKIN BIOPSY

Complications from intradermal injection as well as from the skin biopsy are likely to be minor.

8.10.4 BLOOD, BONE MARROW AND SKIN BIOPSY SAMPLING

This study involves frequent blood and bone marrow sampling. The total number and volume of blood samples is detailed in Table 4.

For immunomonitoring purposes, 70 mL of blood will be collected in heparin coated tubes. Samples will be taken at screening, start vaccination (d=0), day 44, day 77, day 128, day 140 and day 224.

For hematology diagnostic purposes, 21 mL of blood will be collected in heparin coated tubes.

Bone marrow aspirates (at least 3 ml or 16.106 cells for LAP analysis and 10 ml for biobanking) will be collected in heparin coated tubes. Samples will be taken at baseline (d=0), day 98, day 140, and at day 224. All BM samples should be obtained preferably from the first tap, gathered in heparin-coated tubes and kept at room temperature. In any case it should be indicated whether the sample is from the first or second tap. In addition, a single skin biopsy will be taken at 48h post 1st vaccination, 4th vaccination and 2nd booster vaccination. The biopsy will be 4-6 mm and will be taken using a standard punch needle. The injection site to be biopsied will be chosen at random and an injection site will be used for biopsy only once (no repeat biopsy of any injection site).

Table 4: Invasive procedures and sample sizes

Visit no.	1	2	3	4	5	6	7	8	9	10	11	12	13
Day		0	2	14	28	42	44	77	98	126	128	140	224
quantity of Blood (mL)													
For diagnosis	21	21		21	21	21			21	21		21	21
For HLA classification	24												
For immunomonitoring		70	70				70	70			70	70	70
quantity of Bone Marrow aspirate (mL)													
for diagnosis		> 3							> 3			> 3	> 3
For biobank		10							10			10	10
Vaccination													
number of intradermal injections [1]		2-4		2-4	2-4	2-4			1	1			
Skin test													
number of biopsies[2]			1				1				1		

1) Depending on dose: 2 for low dose, 4 for high dose and 1 for boosting dose

2) 1 biopsy to be taken from 1 injection site

9 STUDY OBJECTIVE(S) AND ENDPOINT(S)

The ultimate aim is to reduce the relapse risk through immune-mediated eradication of MRD, thereby prolonging relapse-free survival and overall survival.

9.1 STUDY OBJECTIVE(S)

9.1.1 Primary Objective(s)

1. To assess the effect of DCP-001 on MRD. MRD will be measured by flow cytometry pre and post vaccination as a surrogate marker for established clinical endpoints in AML.
2. To assess the effect of DCP-001 on immune responses in AML patients in first complete remission (CR1) and persistent MRD.
3. To document safety and tolerability.

9.1.2 Secondary Objective(s)

1. To identify surrogate (immunological) markers that might correlate with clinical outcome.
2. To quantify any lag time between initiation of treatment & onset of effect.
3. To determine the effect of DCP-001 on Time to Relapse (TTR).
4. To determine the effect of DCP-001 on Overall Survival (OS).
5. To document the difference between the two vaccination regimens based on MRD outcome and immunological parameters.

9.1.3 Exploratory Objectives

1. To document the number and duration of subsequent remissions.
2. To identify subgroups of patients most likely to benefit from treatment.

Immune monitoring:

Monitoring the immune response is an essential element to evaluate efficacy. The analysis of the immune response against tumour antigens in blood samples from these patients pre and post vaccination allows a better understanding of

1. the underlying mechanisms of action of the immunotherapy;
2. the prognostic value of immunological parameters; and
3. the use of new knowledge about immune parameters to design improved immunotherapy treatments for the future.

The following activities will be performed:

1. Antigen-specific T cell mediated immunity will be measured in PBMC by IFN-α ELISpot and intracellular cytokine release against antigens present in leukemic blasts and also known to be expressed by DCP-001 (i.e. PRAME, WT-1, MUC1, RHAMM). Both pre-and various time points post-vaccination will be analysed.
2. Changes in profiles of different PBMC populations. This includes T-cell profiles, regulatory T cells, MDSC, NK cells etc., according to the same schedule as for antigen-specific responses.
3. General T-cell activation capability and thus immune competence of patients will be analysed by measuring T-cell proliferation and cytokine release following stimulation with DCP-001 in vitro. Pre- and post-vaccination samples will be compared.

4. Biopsies will be taken from the injection site and analysed for specified markers in order to identify the local immune infiltration process.

9.2 ENDPOINTS

9.2.1 Primary Efficacy Endpoint

- The effect of DCP-001 on MRD levels as a surrogate marker for established clinical endpoints in AML.
- To assess the effect of DCP-001 on immune responses in AML patients in first complete remission (CR1) with MRD.
- To document safety and tolerability.

9.2.2 Secondary Efficacy Endpoints

- To identify surrogate (immunological) markers that might correlate with clinical outcome.
- To quantify any lag time between initiation of treatment & onset of effect.
- To determine the effect of DCP-001 on Time to Relapse (TTR).
- To determine the effect of DCP-001 on Overall Survival (OS).
- To document the difference between the two vaccination regimens based on MRD outcome and immunological parameters.

9.2.3 Exploratory Endpoints

- To document the number and duration of subsequent remissions.
- To identify subgroups of patients most likely to benefit from treatment.
- To identify surrogate (immunological) markers that might correlate with clinical outcome.
- To quantify any lag time between initiation of treatment & onset of effect.

9.2.4 Safety endpoints:

- Treatment emergent adverse events (TEAEs) will be collected in first 48 hours after the first vaccination to determine (acute) toxicity due to intradermal injection.
- All TEAEs will be collected during the study period for each patient and the grade of toxicity (CTCAE v.4.0, see Appendix B) and relationship to product determined.
- A DSMB will meet after every 5 patients complete treatment to evaluate safety; written reports will be made of each meeting. Additional details are provided in the DSMB charter.
- All SAEs will be reported by the investigator within 24 hours of the first knowledge. All SAEs will be reported in a timely manner to the appropriate regulatory body, consistent with applicable regulations. Information on relevant SAEs will be disseminated between sites in a timely manner. Processes are described in detail in the study specific safety plan.
- The safety will include all subjects signing ICF and efficacy databases will include only those subjects included in the study who received at least one vaccination.

10 STUDY DESIGN

International, multicentre, open-label proof of concept study without randomization and stratification. Two different dose groups are included. Group 1 consists of 10 patients that will receive 25E6 DCP-001 cells per vaccination with two additional booster vaccinations of 10E6 cells. Group 2 consists of 10 patients who will receive 50E6 DCP-001 cells per vaccination with two additional booster vaccinations of 10E6 cells.

11 STUDY POPULATION

11.1 NUMBER AND TYPE OF SUBJECTS

Twenty patients (male or female) with a confirmed diagnosis of AML that are in CR1 or CRi with MRD, greater than 18 years of age, will be enrolled in this study. Patients will be identified and invited to participate by the investigators at the respective study centres.

11.2 INCLUSION CRITERIA

Patients will be eligible for enrolment if the following criteria apply:

1. Confirmed diagnosis of AML according to WHO2016 criteria, including cytological, molecular and cytogenetic criteria (except acute promyelocytic leukaemia/APL).
2. In CR1 or CRi documented by bone marrow examination up to one month before vaccination; CR defined as less than 5% blasts in normo-cellular bone marrow, ANC >1*10E9/L, platelet count >100*E9/L, no evidence of extra-medullary disease. Patients in CRi (patients with <5% blasts but with incomplete blood count recovery) should have platelets >50 E9/L.
3. MRD as defined by multicolour flow cytometry (MFC) at a value of > 0.1%, or detection of specific molecular abnormalities such as NPM1 mutation
4. Patients that are in CR1 or CRi. Patients not having undergone consolidation therapy must have been in CR1 for at least 1 month prior to enrolment.
5. Expected to be willing and able to undergo all study procedures, including outpatient evaluations for clinical and immunological monitoring.
6. Male or female ≥ 18 years of age.
7. Women of childbearing potential must be using anti-conceptive therapy or use two (2) barrier contraceptive methods (one by each partner and at least one of the barrier methods and one must include spermicide, unless spermicide is not approved in the country or region). See section 12.8 for birth control methods deemed acceptable for this study
8. ECOG (WHO) performance status 0-2.
9. Willing and able to provide written informed consent for participation in the study.

11.3 MAIN EXCLUSION CRITERIA

1. APL (M3) type of AML.
2. Patients who have undergone or are scheduled/eligible for allogeneic stem cell transplantation.
3. History of previous allogeneic bone marrow or solid organ transplantation.
4. Uncontrolled or serious infections
5. Ongoing immunosuppressive therapy, other than short use of low dose steroids, i.e. equivalent to an average dose of ≤10mg of prednisone/day.
6. Chemotherapy and antineoplastic hormonal therapy within 28 days prior to the screening visit.
7. Active autoimmune disease.
8. Inadequate liver function (AST and ALT > 3 x ULN, serum bilirubin >3x ULN).
9. Other active Malignancies within the last 5 years, except for adequately treated carcinoma in situ of the cervix or squamous carcinoma of the skin or adequately controlled limited basal cell skin cancer.
10. Pregnant or lactating females.

11. Major surgical procedure (including open biopsy) within 28 days prior to the first study treatment, or anticipation of the need for major surgery during the course of the study treatment.
12. Uncontrolled hypertension (systolic > 150 mm Hg and/or diastolic > 100 mm Hg) or clinically significant (i.e. active) cardiovascular disease.
13. Evidence of any other medical conditions (such as psychiatric illness, physical examination or laboratory findings that may interfere with the planned treatment, affect patient compliance or place the patient at high risk from treatment-related complications.
14. Known HIV, Hepatitis B and/or Hepatitis C infections.
15. History of hypersensitivity to the investigational medicinal product or to any excipient present in the pharmaceutical form of the investigational medicinal product.

11.4 DEFINITION OF SUBJECTS WHO ARE ENTERED INTO THE STUDY

A patient will be defined as having entered the study when he/she has signed/given informed consent. All entered patients will be assigned a subject number.

Patients who do not meet all the eligibility criteria will be considered as “screening failures”. The only screening data to be entered in the eCRF for all screening failure patients will be informed consent date, demographics, eligibility criteria and reason for discontinuation.

12 TREATMENTS TO BE ADMINISTERED

12.1 Study Drug

12.1.1 Investigational Product(s)

DCP-001 vaccine is presented as a direct injectable solution containing a mature dendritic cell preparation suspended in a cryopreservative, Cryostor^{tm}. The vaccine is gamma irradiated and packed in Daiko Crystal Zyneth break-resistant containers. The format is based on vials with a rubber stopper and aluminium crimp seal with flip off cap. The vaccine will be administered intradermal using a 1 mL syringe and a needle (Terumo, 21 GX 1 ½ “, 0.8 x 40mm). The needle and syringe required for injection purposes will be provided as part of the ‘treatment pack’.

12.1.2 Comparative Drug(s)

Not Applicable

12.2 METHOD OF ASSIGNING SUBJECTS TO TREATMENT GROUPS

First 10 patients will receive the lowest dose (25E6cells/vaccination), the 2nd 10 patients will receive the high dose (50E6cells/vaccination).

12.3 DOSE LEVELS

Patients will receive 25E6 DCP-001 cells per vaccination (Group 1; 10 patients) or 50E6 DCP-001 cells (Group 2; 10 patients) per vaccination. Both groups will receive two additional booster vaccination of 10E6 cells.

The first treatment for both dose groups is given at Day 0 (Baseline), the second, third and fourth treatments are given at 2 weekly intervals. A total of 4 treatments will be administered in a 6 week timeframe followed by a booster vaccination at 8 and 12 weeks after the 4th vaccination at week 6. A summary of the treatment schedule is given below:

Groups 1 and 2;

Day 0	1st vaccination with 25E6 or 50E6 cells.
Week 2	2nd vaccination with 25E6 or 50E6 cells.
Week 4	3rd vaccination with 25E6 or 50E6 cells.
Week 6	4th vaccination with 25E6 or 50E6 cells.
Week 14 and 18	booster vaccination with 10E6 cells.

12.4 ROUTE OF ADMINISTRATION

Intradermal (i.d.) administration of the DCP-001 vaccine will be performed by an experienced and suitably trained nurse or physician. The first vaccine is given at Day 0. Two (25E6) or four (50E6) intradermal injections of 0,5 mL max into the upper left or right leg. The second, third and fourth DCP- 001 vaccinations are given at bi-weekly intervals, in close proximity to the first injections. A booster vaccination will be administered on Day 77 (visit 8) and Day 98 (visit 9).

After each vaccination, patients should be monitored for at least 60 minutes to exclude any immediate reactions to the intradermal administration.

Subjects can choose to alternate the leg the vaccine is administered in, or they can have the vaccine in the same leg throughout the study.

Training on vaccine administration and location of injections will be given to all study site members responsible for vaccine administration prior to the start of the study.

12.5 END OF TREATMENT

Study discontinuation refers to complete withdrawal from the study treatment. The reason for discontinuation from treatment must be recorded. Study discontinuation can be based on one or more of the following criteria:

- Adverse event(s) (physician's discretion)
- Disease progression
- Protocol violation
- Subject withdrew consent
- Lost to follow-up
- Administrative problems
- Death
- Treatment duration completed as per protocol

Furthermore, treatment ends when:

1. No return to baseline or no return to grade 1 non-hematological toxicity at any time during/after the vaccination.
2. Related SAE not otherwise categorized within the CTC v4.0 criteria (see Appendix B) but defined as severe or life threatening at any time during/after vaccination.

Based on the above, there is a differentiation between treatment withdrawal (withdraw treatment, but continue follow-up) and study drop-out (withdrawal from study e.g. unwillingness of the patient to comply with study procedures, withdraw consent, or lost to follow-up).

DCPrime must be notified and the reason(s) for treatment withdrawal/study drop-out should be documented in the eCRF; all follow-up data should also be reported.

Patients who have received at least one dose of DCP-001 who are withdrawn, or withdraw from the study will be treated according to protocol as EOT and EOT pages will be completed. If the patient does not withdraw informed consent, he/she will be requested to enter the follow up phase of the study. These patients will follow the normal schedule of the study excluding the injection of study treatment and the procedures at these visits will occur at the investigators' discretion. These patients will continue to be followed up until EOS or EOT of last patient who completed the study, whichever comes first

If the patients who are required to enter follow up do not enter follow up, for any reason, they must be requested to complete an EOT and EOS visit.

12.5.1 SUBJECT REPLACEMENT

Patients who discontinue due to an (S)AE must be followed-up with appropriate clinical and/or laboratory tests until the (S)AE has resolved satisfactorily, in the opinion of the investigator.

If a patient discontinues for non-(S)AE reasons at any time after entering the study, but before receiving the first treatment, the patient will be replaced.

Patients who discontinue from study treatment within 2 months after start of study treatment for other reasons than ADR(s) will be replaced. Patients withdrawing for ADRs will NOT be replaced. Study drop- outs can be replaced if needed.

12.6 PRIOR AND CONCOMITANT THERAPY

All prior and concomitant medications/therapies will be collected from 28 days before the screening visit and throughout the study, and will be recorded in the eCRF. The exclusion criteria list the prior and concomitant medications/therapies that affect study eligibility.

Throughout the study, all medications deemed necessary for the patient's welfare and are not expected to interfere with the evaluation of the study drug may be administered at the investigators' discretion. If investigators have any questions regarding the suitability of a particular concomitant medication, the Coordinating Investigator should be contacted.

Concomitant therapy with other investigational drugs is forbidden. No immunosuppressive treatment may be offered such as cyclosporine, cyclophosphamide, methotrexate and others except for short use of low dose prednisone (up to a maximum of 10mg/day or other corticosteroid equivalent dose) if indicated for example for concomitant dermal diseases such as Sweet syndrome and/or eczema.

12.7 BIRTH CONTROL METHODS

Patients who are able to become pregnant during the study period should be advised to use appropriate contraception methods. As highly effective birth control methods the following are to be advised:

- Combined (estrogen and progestogen) containing hormonal contraception (oral, intravaginal or transdermal)
- Progestogen-only hormonal contraception (oral, injectable or implantable)
- Intrauterine device (IUD)
- Intrauterine hormone-releasing system (IUS)
- Bilateral tubal occlusion
- Vasectomized partner
- Sexual abstinence

13 STUDY DRUG MANAGEMENT

13.1 PACKAGING AND LABELLING

The investigational drug product DCP-001 will be manufactured at Apceth GmbH, a qualified licensed manufacturer specialized in production of cell therapeutic products. Manufacturing records and certificate of analysis will be issued for all batches to allow release and certification by a Qualified Person (QP). Release by the QP will be performed in accordance with Good Manufacturing Practice (GMP) guidelines, ICH GCP guidelines and applicable local laws/regulations.

DCP-001 is presented as a direct injectable solution containing a dendritic cell preparation suspended in Cryostor™ and stored frozen at -≤135°C. The vaccine is packed in Daiko Crystal Zenith vials. These vials are USP Class VI vials, Sterile (SAL 10-6) according to ISO 11137. The vial format is based on the traditional glass vial with a rubber stopper and aluminium crimp seal with flip-off cap. This will allow easy and sterile bedside handling of the injectable DCP-001.

Sample Label:

DCP-001 1,25 ml Cell concentration see COA Zellkonzentration siehe Analysenzertifikat
Sponsor: DCPrime BV, Galileiweg 8, NL-2333 BD Leiden Advance II Study
CRO: Linical Accelovance, xxxxxxxxxxxxx, xxxxxxxxxxx EudraCT-Nr.: 2017-002869-22
Cell suspension for intradermal application Zellsuspension zur intradermalen Anwendung
For clinical trial use only Nur zur klinischen Prüfung
Store at / Lagerung bei ≤ -135°C Lot/Ch.-B.: DC000XX Exp / Verwendbar bis: MM JJJJ

13.2 STUDY DRUG HANDLING AND STORAGE

Upon receipt at the clinical site, the DCP-001 vials should be immediately transferred to a ≤-135 °C freezer. At the time of vaccination, the vials are removed from the freezer, placed on dry ice, thawed in a 37° water bath according to written procedures, and drawn into the labelled syringes. The syringes are then placed on wet ice, or in a refrigerated box, and released to the appropriated personnel for intradermal injection. Primed syringes must be kept on wet ice or in a refrigerated box until immediately before the vaccine is administered. All injections must be given within 60 min of end of thaw cycle.

13.3 RESPONSIBILITIES FOR STUDY DRUG(S)

Current ICH GCP guidelines require the investigator to ensure that study medication deliveries from the sponsor are received by a responsible person (e.g. a pharmacist), and

- that such deliveries are recorded
- that study medication is handled and stored safely and in accordance with the pharmacy manual
- that study medication is only dispensed to study patients in accordance with the protocol
- that any unused study medication is to be returned to the sponsor.

Drug inventory and accountability records for the medication in this study will be kept by the pharmacist. The following guidelines are therefore pertinent:

- The investigator agrees not to supply study medication to any person except the patients in this study.
- The pharmacist will keep the study drugs in a pharmacy or other locked and secure storage facility, accessible only to those authorized by the investigator to dispense these investigational drugs.
- A study medication inventory will be maintained by the pharmacist. The inventory will include details of materials received and a clear record of when they were dispensed and to which patient.
- At the conclusion or termination of this study, the pharmacist agrees to conduct a final drug supply inventory and to record the results of this inventory on the Drug Accountability Record.
- Unused study medication or medication not dispensed will be returned to the sponsor. It must be possible to reconcile delivery records with those of used and returned medication. Account must be given for any discrepancies. Certificates of deliveries and returns must be signed by the responsible person and made available to the Sponsor, or their representative.

The procedures to ensure appropriate storage, handling and accounting of study medication, will be presented in the Pharmacy Manual supplied to each clinical centre pharmacist or delegate.

As this study involves intradermal administration of the study medication administered by trained study personnel, patient compliance measures are not necessary. Study medication will be administered under direct supervision, according to the Pharmacy Manual and the clinical centre's standard operating procedures.

14 STUDY PROCEDURES

14.1 STUDY PROCEDURES AT EACH VISIT

Patients will be screened for eligibility to participate in the study within 4 weeks prior to the first vaccination.

Visit 1 (Screening Visit)

Screening assessments will include:

- Obtaining of written informed consent prior to any study specific procedure (including screening procedures) being performed
- Verify all inclusion criteria are met and no exclusion criteria are met
- Record demographics
- Complete medical history; including previous therapies for AML
- Perform physical examination assessment. The physical examination of baseline will also include WHO Performance status (ECOG)
- Record vital signs (heart rate, blood pressure and oral body temperature)
- Urine analysis
- Perform study specific haematology and biochemistry (incl. beta hCG blood test in woman with child bearing potential)
- Viral screen (HIV, Hepatitis B and C)
- ECG, X-thorax
- HLA typing
- Record any prior and concomitant medications

All laboratory procedures (including central laboratory) will be described in the Laboratory manual.

Treatment Period; Visit 2 (Baseline Visit)/ day 0

The Baseline Visit will include:

- Physical examination, ECOG and Vital signs
- Perform study specific hematology and biochemistry
- Record any new AEs, SAEs and concomitant medications and/or changes in ongoing AEs, SAEs and ongoing concomitant medications
- Blood sample collection for immunological response (immunomonitoring) before vaccination
- Administration of study medication (DCP-001 vaccination)
- Bone marrow aspirate for MRD analysis (MRD assessment) and biobanking

Treatment Period (Visit 3)/ day 2

- Skin biopsy from 1 out of the 2 or 4 injection sites for evaluation of immunological response in skin biopsy
- Blood sample collection for immunological response
- Record new any AEs, SAEs and concomitant medications and/or changes in ongoing AEs,

SAEs and ongoing concomitant medications
- Day is fixed with the previous visit (Visit 2)

Treatment Period (Visit 4-6)/days 14, 28 and 42

The following assessments will be performed at the indicated visits according to the assessment schedule ± 3 days.

- Physical examination, ECOG and Vital signs
- Hematology and biochemistry
- Record any AEs, SAEs and concomitant medications and/or changes in ongoing AEs, SAEs and ongoing concomitant medications.
- Administration of study medication (DCP-001 vaccination)

Treatment Period (Visit 7)/ day 44

- Skin biopsy from 1 out of the 2 or 4 injection sites for evaluation of immunological response in skin biopsy.
- Blood sample collection for immunological response
- Record any AEs, SAEs and concomitant medications and/or changes in ongoing AEs, SAEs and ongoing concomitant medications
- Day is fixed with the previous visit (Visit 6)

Treatment Period (Visit 8)/ day 77

according to the assessment schedule ± 3 days

- Physical examination, ECOG and Vital signs
- Record any new AEs, SAEs and concomitant medications and/or changes in ongoing AEs, SAEs and ongoing concomitant medications
- Blood sample collection for immunological response

Treatment Period (Visit 9-10)/days 98, 126

according to the assessment schedule ± 3 days
- Physical examination, ECOG and Vital signs
- Hematology and biochemistry
- Record any new AEs, SAEs and concomitant medications and/or changes in ongoing AEs, SAEs and ongoing concomitant medications.
- Administration of study medication (DCP-001 booster vaccination)
- ***Visit 9;*** Bone marrow aspirate for MRD analysis (MRD assessment) and biobanking

Treatment Period (Visit 11)/day 128

- Skin biopsy from 1 out of the 2 or 4 injection sites for evaluation of immunological response in skin biopsy
- Blood sample collection for immunological response (immunomonitoring)
- Record any new AEs, SAEs and concomitant medications and/or changes in ongoing AEs, SAEs and ongoing concomitant medications
- Day is fixed with the previous visit (visit 10)

Treatment Period (Visit 12)/day 140

according to the assessment schedule ± 3 days

- Physical examination, ECOG and Vital signs
- Hematology and biochemistry
- Record any AEs, SAEs and concomitant medications and/or changes in ongoing AEs, SAEs and ongoing concomitant medications.
- Bone marrow aspirate for MRD analysis (MRD assessment) and biobanking
- Blood sample collection for immunological response (immunomonitoring)

Treatment Period (Visit 13)/ day 224

according to the assessment schedule ± 3 days

- Physical examination, ECOG and Vital signs
- Hematology and biochemistry
- Record any new AEs, SAEs and concomitant medications and/or changes in ongoing AEs, SAEs and ongoing concomitant medications.
- Bone marrow aspirate for MRD analysis (MRD assessment) and biobanking
- Blood sample collection for immunological response

Follow-up visits (14-17)

Upon completion of the treatment phase each patient will enter the follow up phase. The patient will return every 8 weeks (± 2 weeks) up to 12 months after 4th vaccination. The following assessments will be performed at each follow up visit;

- Physical examination, ECOG and Vital signs
- Hematology and biochemistry
- Record any new AEs, SAEs and concomitant medications and/or changes in ongoing AEs, SAEs and ongoing concomitant medications

Subsequent follow up visits will occur at least every 8 ± 2 weeks until end of study (i.e. 12 months after 4th vaccination) or disease progression.

End of study visit (18) / 12 months after last vaccination

- Physical examination, ECOG and Vital signs
- Perform study specific hematology and biochemistry
- Urine analysis
- Record any new AEs, SAEs and concomitant medications and/or changes in ongoing AEs, SAEs and ongoing concomitant medications

See schedule of assessment (Table 1) for study specific procedures.

14.2 STUDY EVALUATIONS

14.2.1 Clinical response evaluation

For evaluation of clinical responses, appropriate measures to accurately assess the presence of leukemic blasts in bone marrow or blood will be performed at indicted time point throughout the study (Table 1).

MRD will be assessed in bone marrow samples and Leukaemia Associated Immunophenotypes (LAP) will be established at start of the protocol. These cells will be followed during treatment and the MRD will be reported as %LAP positive cells of the WBC.

14.2.2 Assessment of MRD

MRD will be monitored in bone marrow aspirates by multiparameter flow cytometry assay according to standardized procedures. Bone marrow aspirate samples (at least 3 ml or 16.10E6 cells) will be collected in heparin coated tubes and will be send directly to the central lab at VUmc according to predefined and established procedures (Described in the Laboratory Manual). Samples will be processed according to standardized operating procedures and analysed using standardized gating strategies.

An additional bone marrow aspirate sample of 10 mL, taken at the same time as the analysis sample, will be biobanked according to local biobank laws. Materials will be stored as additional MRD analysis may appear to be relevant at a later stage. Which analyses to be performed will be decided later and will be dictated by the MRD analysis outcome. Samples will only be biobanked if the patients have consented to biobanking.

14.2.3 Assessment of Safety

Safety and tolerability will be assessed throughout the study starting from the time the patient signed the informed consent form until their participation in the study is finalized.

The safety parameters assessed throughout the study include physical examinations, vital signs, ECG recordings, laboratory evaluations (hematology, biochemistry, urine analysis) and recoding of Adverse Events. Throughout the duration of the study, adverse events and concomitant treatments will be recorded continuously.

14.2.3.1 Physical Examination

A general physical examination, including ECOG status, height (only at the screening visit) and body weight, will be performed during the study at different visits according to the assessment schedule presented in the study flowchart (Table 1). All laboratory results and assessment of abnormal results (clinical significance) will be recorded in the eCRF. The physical examination will include the following: head and neck, eyes, ENT (ears, nose, throat), heart, longs, chest, abdomen, skin, musculoskeletal, neurological, endocrine and lymphatic system.

The physical examination performed at the screening visit will also include the ZUBROD-ECOG-WHO Performance status (see APPENDIX A) and the result will be recorded in the eCRF.

Vital signs measurements will be performed after at least 5 minutes of supine rest and will be recorded in the eCRF.

14.2.3.2 Electrocardiogram

ECG recordings will be performed at the screening visit. ECG recording parameters, and investigator assessment will be recorded in the eCRF.

14.2.3.3 Chest X-ray

A postero-anterior chest X-ray will be performed at the screening visit. Only abnormal clinically relevant findings will be recorded in the eCRF.

14.2.3.4 Laboratory Evaluations

Laboratory evaluations (hematology, biochemistry, urine analysis) will be performed at the local institute's laboratory. The following laboratory values will be assessed:

- Hematology: Hemoglobin, Ht, erythrocytes, reticulocytes, platelets, WBC and differential
- Blood chemistry: Na, K, creatinine, Ca, P, Mg, bilirubin, AF, GT, ASAT, ALAT, LDH, glucose, APTT, INR, fibrinogen.
- beta hCG blood test in woman with child-bearing potential
- Viral blood test (HIV, Hepatitis B and C)
- Urine analysis (pH, protein, erythrocytes, leucocytes, glucose, ketones, nitrites)

14.2.4 Assessment of Immune responses

DCP-001 vaccination supposed to develop anti-tumour activity in AML patients by inducing functional T-cell responses against tumour-associated peptides (TAA) derived from WT-1, RHAMM, Mucin-1 and PRAME that have shown to be important targets in AML and are presented by DCP-001. Monitoring the immune response is an essential element to evaluate efficacy. The analysis of the immune response against tumour antigens in blood samples from these patients before and after treatment allows a better understanding of 1) the underlying mechanisms of action of the immunotherapy 2) the prognostic value of immunological parameters and 3) the use of new knowledge about immune parameters to design improved immunotherapy treatments for the future. The following activities will be performed.

a. Antigen-specific T cell mediated immunity will be measured in PBMC by IFN-ϒ ELISpot and intracellular cytokine release against antigens present in leukemic blasts and also known to be expressed by DCP-001 (i.e. PRAME, WT-1, MUC1, RHAMM). Both pre-and various time points post-vaccination will be analysed. In those patients that show positive responses, additional intracellular cytokine staining's (ICS) will be performed
b. Changes in profiles of different PBMC populations. This includes T-cell profiles, regulatory T cells, MDSC, NK cells etc, according to the same schedule as for antigen-specific responses.
c. General T-cell activation capability and thus immune competence of patients will be analysed by measuring T-cell proliferation and cytokine release following stimulation with DCP-001 in vitro. Pre- and post-vaccination samples will be compared.
d. Biopsies will be taken from the injection side and analysed for specified markers by immunohistochemistry in order to identify the local immune infiltration process.

The assessment of the immune responses will be performed by a central lab, i.e. Immatics Biotechnology (www.immatics.com). Immatics is a leading cancer immunotherapy company and the global leader in the discovery of novel targets for various types of cancer immunotherapies.

Immunohistochemistry will be performed by DIAPath at the Center for Microscopy and Molecular Imaging (CMMI) at Gosselies, Belgium.

14.2.4.1 Sample collection, shipment and storage

Blood samples will be taken at indicated Visits (Table 1) from all patients included in the trial.

At the clinical site, a 70 mL heparin blood sample will be collected aseptically. Heparin blood is then transported immediately to the lab that performs PBMC isolation and cryoconservation for the site ('PBMC lab'). Trained and released PBMC operators will separate PBMCs (lymphocytes and monocytes) from other blood components (serum, erythrocytes, thrombocytes and granulocytes), count and freeze isolated cells according to Immatics SOP TC-021 within a total time of 8 h. All critical consumables will be provided by Immatics, including PBMC Kits containing components for isolation, freezing and cryopreservation. Labelling stickers allowing unambiguous identification of each cryovial. Isolation and cryopreservation will be documented in PBMC process slips that will be faxed/mailed to Immatics according to SOP TC-021. PBMC samples will be stored at -80°C for a maximum of one week and then transferred to LN2 tanks. Samples are sent to Immatics on dry ice by a trained courier service. Frozen PBMC samples will be transferred into a gas phase liquid nitrogen cryostorage system until in- house assays will be performed. Sample entry is regulated by Guideline GL-ADM003 and SOP IN-008. Immatics' cryostorage system is internally temperature controlled and additionally monitored by an electronical alarm system.

Biopsies (skin) will be taken via punch biopsies from the injection site 2 days after the first vaccination, 4th vaccination, and the second booster vaccination. Tissue samples must be handled as described in Laboratory manual. In short, samples must be fixed during at least 24 hours in a solution of formaldehyde 4% neutral pH (buffered) (maximum 48 hours of fixation). The volume of the fixative solution must be min 20x higher than the volume of tissue. After tissue fixation, samples will be directly sent to the central lab for further processing and immunohistochemistry analysis.

14.2.4.2 IFN-ϒ ELISPOT Assay

Principle

ELISpot assays are single-cell based and result in a parameter (the frequency of spot forming cells among mononuclear cells) that is expected to be correlated to the true frequency of activated antigen specific T lymphocytes in blood samples. As such assays also allow the processing of large numbers of samples and antigens in parallel, they have been widely used for immunomonitoring studies in a large number of clinical trials.

Description

In order to quantitatively determine the IFN-ϒ secretion by PBMCs isolated from peripheral blood samples a precise and accurate IFN-ϒ Elispot assay has been successfully developed and validated during two clinical trials performed by Immatics (IMA901-101 and IMA901-202). The assay is optimized with respect to the pre-sensitization conditions (medium, human serum, antigen concentration, duration), as well as readout conditions (antigen concentration). Thresholds for positive responses are defined by three criteria: 1) t-test (antigen triplicate vs. negative control triplicate), 2) ratio for mean of antigen vs. mean of negative control spots > 2.0 and 3) mean of antigen triplicate of spots per effector cells must be at least 6.0.

Statistical analysis for T-cell responses

Here, criteria for detection of above-threshold responses at given time points in a particular assay are defined. Those T-cell responses may be either vaccine-induced or due to other influences such as activated or naïve T cells prior to therapy.

Criteria for identification of above-threshold T-cell responses from ELISpot data are based on the recommendations of the CIMT Immunomonitoring panel (19). Further criteria increasing the stringency of the CIMT criteria were included to exclude the possibility of detection of false-positive responses.

Criteria for vaccine-induced T-cell responses in amplified Elispot assays

1. Criterion 1: Positive above-threshold immune response in a post-vaccination time point.
2. Criterion 2: For ELISpot assays, Rpost must be at least 2.0x of Rpre. Rpost and Rpre are the ratios at post- and pre-vaccination for the same antigen, patient & assay, for mean of antigen vs. control (triplicate of spots per 150.000 effector cells).

Note 1: If the criterion 2. cannot be calculated due to division by zero, 0.1 spot per 150.000 effector cells is added to all wells of pre-vaccination time point for that patient / assay combination and 2. is recalculated.

Note 2: If the criterion 2. cannot be calculated due to the flag for "too numerous to count", the respective well will be set to 1000 spots per 150.000 effector cells for the same assay and 2. is recalculated.

14.2.4.3 ICS Assay

Principle

The Intracellular Cytokine Staining (ICS) assay has the capability to detect multiple effector functions, including IFN-gamma. It has been shown that such assays allow the detection of several cytokines and other functionalities at the same time on the same cell, and have been suggested as optimal for the sensitive and quantitative assessment of antigen-specific T cells in vaccination trials if multiple effector functions should be measured.

Description

In order to analyse the functionality and therefore the quality of T-cell responses a sensitive, quantitative ICS assay was developed which allows immunophenotyping of responding cells and measurement of multiple effector functions on a single cell level. This ICS assay has been validated during four clinical trials (IMA901-202, IMA910-101, IMA901-301, IMA950-101). The assay is optimized with respect to the pre-sensitization conditions (medium, human serum, antigen concentration, duration), as well as readout conditions (cells per well, antigen concentration, restimulation, antibody titrations and staining conditions). Thresholds for positive responses are defined by 3 criteria: 1) Positivity of at least one functional pattern (T cells expressing one of the functional markers, e.g. IFN- ϒ, TNFα, CD154, IL-10, IL-2, IL-4 or IL-5), 2) Frequency of functional T cells for an antigen must be at least 4x the frequency of the MOCK control for the same functionality and 3) Fisher exact test (two- sided) for counts of functional T cells and non-functional T cells for antigen vs. MOCK control must be $p<0.001$.

Statistical analysis for T-cell responses

Criteria for identification of above-threshold T-cell responses for the ICS assay are based on ELISpot criteria.

Criteria for vaccine-induced T-cell responses in amplified ICS assays

1. Criterion 1: Positive above-threshold immune response in a post-vaccination timepoint.
2. Criterion 2: For ICS assays, Dpost must be at least 4 x Dpre. Dpost and Dpre are the differences of T-cell frequencies at post- and pre-vaccination for the same antigen, patient & assay, for antigen minus HIV-001 control.

14.2.4.4 Treg / MDSC / PROFILE Assays

Principle

Characterisation of immune cell activation and effector phenotypes is an important measure of responses to immunotherapy. DCP-001 vaccination-induced changes in the frequency and/or activation status of different immune cell populations (e.g. T cells, NK-cells, regulatory T cells, Monocytes) will be determined for available patient PBMC, as these may be informative in further dissecting the nature of the response. Flow cytometry will be used to detect and quantify cells stained with fluorescently labelled antibodies. Additionally, other cell populations including but not restricted to regulatory T cells and myeloid-derived suppressor cells shall be determined by multicolour flow cytometry to investigate potential correlations with T cells responding to tumour-associated peptides derived from WT-1, RHAMM, Mucin-1 and PRAME

Description

In order to assess cellular biomarkers three different assays will be used: 1) a Treg assay (analysing regulatory T Cells), 2) an MDSC assays (analysing myeloid-derived suppressor cells) and 3) the PROFILE assay (analysing the profile of cells present in the PBMC compartment). The Treg and the MDSC assay have been developed for a clinical trial of Immatics (IMA901-202) and has been validated during 5 clinical trials. The PROFILE assay has been developed for this trial and will be used for the first time. The Treg and MDSC assays have been optimized in terms of antibody titrations, instrument settings, compensation and isotype controls. Validation has been assessed for intra-assay variability, inter-assay variability and accuracy.

14.2.4.5 Lymphocyte proliferative response

Principle

In addition to antigen specific responses, possible changes in vaccination induced systemic lymphocyte responses will be measured by analysing lymphocyte proliferation in response to co- culture with the vaccine in vitro. This assay serves as a marker for general immune-competence of the patient. Such in vitro proliferative responses of both CD4 and CD8 T cells were monitored in the Phase I study that has been completed with DCP-001, and showed an increase after vaccination in a number of patients.

Description

To assess the immune competence of patients pre and post vaccination, the lymphocyte proliferative response will be evaluated by incubating patient derived fluorescent dye labelled PBMC with graded doses of irradiated stimulator cells (DCOne derived mDC and progenitor cells) in round-bottom, 96- well plates in triplicates for 3-6 days. A positive vaccination-induced response is defined as an increase in Post-Vaccination proliferation of ≥ 10%. Supernatants of all samples will be collected for exploratory cytokine analysis using a luminex bead-based multiplex assay.

14.2.4.6 Immunohistochemistry (IHC) on biopsies

Principle

To obtain more insight in the immune cells infiltrating the injection site, punch biopsies will be taken from the injection site 2 days after the first vaccination, 4th vaccination, and the second booster vaccination. The cellular infiltrate will be analysed by hematoxylin-eosin (HE)staining IHC in order to characterise the inflammatory cell populations by digital quantification that allows quantitative, standardised and calibrated image acquisition.

Description

A hematoxylin-eosin (HE) staining will be performed on two tissue slides located just before and after the last IHC slide. A pathologist will provide a morphological evaluation of the two HE tissue slides. IHC will be performed for each biopsy on serial slide sections. Based on a review of IHC slides by a pathologist either a semi-quantitative analysis (scoring), or a computer-assisted approach (digital image analysis) will be performed.

14.3 PHARMACOKINETICS

Not applicable.

14.4 ADHERENCE TO PROTOCOL

14.4.1 Treatment Compliance

As this study involves intradermal administration of the study medication administered by trained study personnel, patient compliance measures are not necessary. The quantity of study medication administered to the patient will be recorded in the eCRF at each visit.

14.5 TERMINATION OF THE STUDY BY THE SPONSOR

The study may be terminated prematurely in case of the occurrence of serious, unexpected adverse events, necessitating a review of the product's safety profile. Also, unforeseen events or new information relating to the product, making it unlikely that the objectives of the study or clinical programme will be met, may warrant the sponsor to discontinue the study prematurely.

The Sponsor reserves the right to discontinue the study at any time for failure to meet expected enrolment goals and therefore it will not be possible to reach study objectives, in the interests of safety, or any other administrative reasons e.g. in those cases that compliance to the clinical protocol can no longer be guaranteed. The Sponsor shall take advice from the data and safety monitoring committee as appropriate in making this decision.

In case the study is ended prematurely, the sponsor will notify the Relevant Competent Authorities and ECs and will including the reasons for the premature termination.

15 ADVERSE EVENTS AND OTHER SAFETY ASPECTS

15.1 DEFINITION OF AN ADVERSE EVENT

An adverse event (AE) is any undesirable physical, psychological or behavioural effect experienced by a patient during participation in an investigational study, in conjunction with the drug or biologic, whether or not product-related. This includes any untoward signs or symptoms experienced by the patient from the time of signing of the informed consent until completion of the study.

AEs may include, but are not limited to:

- subjective or objective symptoms spontaneously offered by the patient,
- and/ or observed by the investigator or medical staff,
- findings at physical examinations,
- laboratory abnormalities of clinical significance.

Disease signs, symptoms, and/or laboratory abnormalities already existing prior to the use of DCP-001 are not considered AEs after treatment unless they reoccur after the patient has recovered from the pre-existing condition or in the opinion of the investigator they represent a clinically significant exacerbation in intensity or frequency.

Clinical significance is defined as any variation in signs, symptoms, or testing that has medical relevance and may result in an alteration in medical care. The investigator will continue to monitor the patient until the assessment returns to baseline or until the investigator determines that follow-up is no longer medically necessary.

Worsening of the patient's condition for which the study treatment is being used is not considered an AE.

15.2 DEFINITION OF A SERIOUS ADVERSE EVENT

A serious AE (SAE) is any AE that results in the following outcomes:

- results in death,
- life threatening experience,
- required or prolonged inpatient hospitalization,
- persistent or significant disability/incapacity,
- congenital anomaly,
- important medical events, that based upon appropriate medical judgement, may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed above.

15.3 ADVERSE EVENT REPORTING

The investigator is responsible for monitoring the safety of patients who have enrolled in the study and for accurately documenting and reporting information as described in this section.

Patients will be instructed to report to the investigator any AE they experience. Investigators will ask about the occurrence of AEs at each visit. Investigators are required to document all AEs occurring during the clinical study, commencing with signing of the informed consent through the protocol defined end of study.

Adverse events will be scored according to the NCI Common Terminology Criteria for Adverse Events, version 4.0.

The causal relationship of AEs to study treatment will be determined by the Investigator and adjudicated by the MM. Drug-related causality will be defined as events that are definitely related, probably related, or possibly related to study drug. Events will be categorized as unrelated to study drug if they are reported as not related. The following list will be used to assist with the assessment of causality:

Definitely Related: The AE is clearly related to the investigational agent/procedure, i.e., an event that follows a reasonable temporal sequence from administration of DCP-001, follows a known or expected response pattern to the suspected drug, that is confirmed by improvement on stopping and reappearance of the event on repeated exposure and that could not be reasonably explained by the known characteristics of the subject's clinical state.

Probably related: When there is a temporal relationship to the study drug administration, significant symptoms abate upon discontinuation of DCP-001 and there is a reasonable explanation based on known characteristics of the study drug and there is no clear association with pre-existing disease or therapy, intercurrent illness, concurrent therapy, or other factor(s).

Possibly related: An event that follows a reasonable temporal sequence from the administration of DCP-001, follows a known or expected response pattern to the suspected drug, but that could readily have been produced by a number of other factors.

Not related: The AE is clearly not related to the investigational agent/procedure. Another cause of the event is most plausible; and/or a clinically plausible temporal sequence is inconsistent with the onset of the event and the study medication administration; and/or a causal relationship is considered biologically implausible. Concurrent illness, concurrent medication or other known cause is clearly responsible for the AE.

The action taken with study treatment will be reported according to the following options: Dose not changed, Dose reduced, Drug interrupted, Drug withdrawn, Not applicable, and Unknown.

In accordance with international regulatory requirements, there is a separate reporting system for SAEs.

15.4 DOCUMENTATION OF NON-SERIOUS ADVERSE EVENTS

All AEs occurring following the signature of the informed consent will be recorded. All AEs will be recorded in designated eCRF pages and source medical recorded.

15.5 REPORTING OF SERIOUS ADVERSE EVENTS

In case of a SAE, the investigator should contact Linical Accelovance, by telephone, email or fax within 24 hours of the first knowledge of the event even if the experience does not appear to be related to the study drug (contact details to be used: Linical Accelovance Drug Safety (using the SAE Cover Sheet Report Forms – in the SAERP) to Linical Accelovance's SAE dedicated fax#: 1-866- 857-8839 or SAE@linical.accelovance.com. See also page 8.

Full details of the SAE should be recorded on the observation notes and on the eCRF.

The investigator must also complete an SAE Report, containing all information that is required by Regulatory Authorities. The completion of this SAE Report is therefore mandatory for SAEs.

All correspondence on SAEs has to be marked URGENT. The following minimum information is required:

- Study number
- Patient identification number, sex, and year of birth
- The name and address of the reporting person (in confidence)
- A description of the SAE

The sponsor will report expedited the serious unexpected serious adverse reactions (SUSARs) to the IEC:

- SUSARs that have arisen the clinical study that was assessed by the IEC;
- SUSARs that have arisen in another clinical study of the same sponsor and with the same medicinal product, and that could have consequences for the safety of the patients involved in the clinical study that was assessed by the IEC.

The remaining SAEs will be reported to the IEC on half yearly basis in form of line listings, accompanied by a brief report highlighting the main points of concern.

The sponsor will report all SUSARs on expedited basis to the competent authorities.

The expedited reporting will occur not later than 15 calendar days after the sponsor has first knowledge of the adverse reactions. For fatal or life threatening cases the term will be a maximum of 7 calendar days for a preliminary report with another 8 days for completion of the report.

15.6 EXPECTED SIDE EFFECTS OF STUDY MEDICATION

Results from numerous clinical trials with DCs indicate that DC-based immunotherapy is safe and well-tolerated. There may be a slight discomfort due to the local skin reaction following injection.

15.7 FOLLOW-UP OF CLINICALLY SIGNIFICANT ADVERSE EVENTS

All AEs will be followed until they have abated, or until the investigator determines that follow-up is no longer medically necessary. Depending on the event, the follow up may require additional tests or medical procedures as indicated, and/or referral to the general physician or medical specialist.

15.8 DATA SAFETY MONITORING BOARD

The DSMB will meet per the DSMB Charter.

The primary responsibility of the Data Safety Monitoring Board (DSMB) is to ensure safety of patients prior to administration of the next vaccination of DCP-001. The DSMB consist of qualified experts in the disease area but are otherwise independent of the study. In case of doubt regarding the interpretation of any AEs, responsible investigators will on the shortest notice report the issue to the medical monitor who will consult with the DSMB, at least in case of any SAE determined to be related to DCP-001 administration. The DSMB will review each SAE and all severe laboratory abnormalities as rapidly as possible and provide advice whether the study can reasonably continue. Other AEs, including laboratory data which may indicate toxicity, will be reviewed periodically. The DSMB will discuss, analyse on the issue, and report to the investigator and the sponsor. Ultimately the sponsor will decide whether the study should be discontinued (section 14.6).

16 STATISTICAL METHODS

16.1 DETERMINATION OF SAMPLE SIZE

Recruitment will target 20 patients greater than 18 years of age with a confirmed diagnosis of AML that are in CR1 or CRi with MRD. The planned sample size of 20 evaluable subjects will be distributed 1:1 with 10 subjects each in Group 1- 25E6 DCP-001 cells per vaccination, and Group 2- 50E6 DCP-001 cells per vaccination. Additional subjects may be enrolled to ensure that 20 evaluable subjects complete the study. As the impact of vaccination on MRD levels is the primary endpoint, strict sample size statistical considerations with a desired power for a pre-specified difference is not yet possible. However, assuming that 90% of patients in the overall (untreated) population will experience MRD, a sample size of 20 subjects is sufficient to detect a 22% difference between the treated group and the overall population, using a one sample, two-tailed z-test of proportion with 80% power and a 5% level of significance. No formal sample size calculations were completed for this study.

16.2 STATISTICAL AND ANALYTICAL PLANS

No inferential analysis is planned for the study; only descriptive analyses will be used. In general, descriptive statistics, unless otherwise noted, will include the number of subjects, mean, standard deviation, median, minimum value, and maximum value, and where applicable, 95% confidence intervals. Percentages will be calculated using the number of evaluable subjects within each treatment group. If appropriate, immune monitoring data will be summarized using the geometric mean, standard error of the geometric mean, and 95% confidence intervals of the geometric mean. Unless otherwise stated, all summary tables will present descriptive statistics and/or frequency by visit for each treatment group. The analysis will be performed on the data collected up to the last MRD assessment (Day 224).

All statistical analyses and data summaries will be completed using SAS Software (version 9.2 or greater).

16.2.1 Definition of Analysis Population

The "Safety Population" will be comprised of all subjects who provide informed consent and receive at least one vaccination. Subjects in the Safety Population will be analysed as treated.

The “Immunogenicity Population” will be comprised of all subjects who received a vaccination and for whom any follow-up immune monitoring data were recorded. This is the dataset to be used for the primary outcomes.

16.2.2 Handling of Missing Data

No adjustments in the analysis for missing data are planned. However, missing immune monitoring data will be explored and, if significant, appropriate statistical methods may be applied to adjust for missing data.

16.2.3 Analysis of Efficacy Variables

MRD levels and immune responses will be summarized descriptively by treatment group and visit as described above. Time to event variables (e.g. time to relapse) will be analysed using survival analysis techniques (e.g, Kaplan-Meier estimates).

16.2.4 Safety analyses

AEs will be classified as treatment emergent AEs (TEAEs) if they start on or after the first vaccination, through 30 days after the last vaccination. AEs that are present prior to the first vaccination, but their severity or relationship increases after the first vaccination, will also be classified as TEAEs.

TEAEs will be tabulated using MedDRA body system and preferred term. The number and percentage of subjects experiencing at least one TEAE overall and at least one TEAE for a body system and preferred term will be tabulated by toxicity grade (CTCAE) and treatment group. A similar summary will be completed by relationship to study product and treatment group. For the calculations in these tables, each subject's TEAEs will be counted once under the maximum toxicity grade or the strongest recorded causal relationship to study product. A summary will be performed for all serious TEAEs. A listing of SAEs will provide details of the events including severity, relationship to study product, time between onset and last vaccination, number of vaccinations received, and a summary of the event. A summary and listing will also be presented for TEAEs causing treatment discontinuation.

Clinical Laboratory results will be summarized descriptively for each treatment group by visit for the observed value as well as for the change from baseline value. In addition, laboratory shift tables will be provided for all laboratory parameters where low/normal/high or abnormal/normal status can be ascertained. In the event of repeat values, the last non-missing value per study day/time will be used for summarization. Listings of individual laboratory parameters by visit with normal ranges and abnormality assessments will also be completed by subject. Laboratory listings will include the study day, date collected, and observed laboratory values with out-of-range values flagged. Unscheduled visit laboratory results will not be summarized but will be included in patient data listings.

For vital signs, descriptive statistics of observed values will be presented for each scheduled visit by treatment group. Changes from baseline to each scheduled post-baseline visit will be presented. All vital sign data by subject will be presented in a listing. Unscheduled visit vital sign data will not be summarized but will be included in patient data listings.

The number and percentage of discontinuation of vaccination and early study termination due to various reasons will be tabulated by treatment group and overall.

Other safety data (e.g. vaccine administration, physical examination, concomitant medications, ECG) will be listed, and summaries presented if appropriate.

Full details of all summary and analysis will be provided in the statistical analysis plan.

16.3 RANDOMISATION

Not Applicable

First 10 patients will receive the low dose (25E6 cells/vaccination followed by 2 booster vaccinations), followed by 10 patients receiving the high dose (50E6 cells/vaccination)

17 DATA MANAGEMENT AND QUALITY ASSURANCE

17.1 DATA COLLECTION

The Investigator will complete and maintain source documents for each subject participating in the study. Which documents will be regarded as source data will be agreed with the site and documented on a Source Documentation Log at the SIV. All information required by the protocol should be documented in the source records. An explanation must be given for any omissions.

Source data must be available at each site visit to document the existence of the study subjects and substantiate integrity of study data collected. Source data must include the original documents relating to the study, the medical treatment and medical history of the patient.

The following information will be included in the source medical records:

- Demographic data
- Details related to the inclusion criteria
- Informed consent forms
- Medical history and physical examination details
- Adverse events and concomitant treatment(s)
- Results of relevant examinations (e.g. ECG traces, X-ray etc.)
- Laboratory printouts
- Visit dates and dispensing of study medication

Data of this study will be recorded via electronic data capture (EDC) in an eCRF. Data collected on each study patient will be recorded on the eCRF. The eCRF is part of a system for the capture and management of the data, fully validated and 21 CRF part 11 compliant. The CRO personnel will train designated investigator study centre staff in the eCRF. Investigator study centre staff will not be given access to the eCRF until they have been trained. Only authorized study staff will enter data required by the study protocol in the eCRFs. The investigator must certify that the data entered in the eCRFs are complete and accurate by signing off on the eCRF forms. After database lock, the investigator will receive a CD-ROM with the eCRFs of the patient data for archiving at the investigational study centre.

17.2 STUDY MANAGEMENT

A Project Manager (PM) will be assigned to manage the clinical activities. The PM will be responsible for managing the project team and will provide the liaison activities with the project team, the CRA's, the Medical Monitor and any regulatory representatives as necessary. The PM will work toward the identification and resolution of any problems and will coordinate the daily project management operations. This task involves management and document development related activities (i) before the study start up, (ii) upon start up and (iii) during the studies.

i. Before the study start up, participating sites will be prepared and assessed. The team will conduct an on-site pre-study Site Qualification Visit (SQV) at all sites. This is followed by signing of the Clinical Trial Agreement (CTA) by the main investigators and mutual agreement on contracts and budgets. The CRO team, in collaboration with the sponsor, will generate presentation materials for site initiation and the site's Regulatory Binder and Essential Documents (ICH-GCP (E6)) will be prepared. Other project documents that will be prepared for implementation in the studies are

the Project Management Plan Monitoring & Communication Plan, Source Document Log Templates and Monitoring Visit Report templates and new SOP's as may be required.

ii. Upon the study start, the clinical team members will be trained on procedures (including clinical sample management) and oriented on documents and data management. Also the project team, including CRAs, will be trained on project-specific documents and relevant SOPs. Project management and monitoring tools and tracking systems will be prepared to implement on the sites.

iii. During the studies, the project infrastructure, project documents and project files will be maintained and updated as needed. The PM will facilitate the recording and distribution of meeting minutes for project team meetings so that relevant information is communicated to the project team.

iv. The PM team will be responsible for creation, maintenance and transfer of the Trial Master File (TMF).

17.3 STUDY MONITORING

The trial will be monitored according to applicable ICH, GCP and regulatory guideline, preparation of data queries and reports to assist the clinical monitoring, user acceptance test plans, data review checklists, database quality acceptance sampling plans, database lock checklist sign-off sheet and training each study site staff member entering data in the eCRF.

Clinical Research Associates (CRA's) will provide site monitoring activities and a Medical Monitor (MM) to keep medical oversight of the trial procedures involving communication with the main investigators on subject inclusion, coordination of protocol deviations or other changes and support to sites and project teams.

The CRAs will travel to each of the sites and conduct an on-site initiation visit (1 per site) to present initiation materials, confirm that all staff understands the requirements for the study and that all processes and materials required for the trial are present. The CRAs will answer questions that arise during the meeting, and generate and provide a follow-up letter to the sites for their study files.

The scheduled number of routine/interim monitoring visits (5 per site) is required to evaluate site compliance with the protocol and data for inclusion. Additional visits or additional days on site may be required to address additional workload or questions of quality or compliance, for site retraining in the event of staffing changes or excessive or major protocol deviations.

Monitoring visits will be scheduled and conducted according to the Monitoring Plan. Each visit will be documented in an Interim Monitoring Visit report (IMV).

Routine site management and communication is done by CRAs according to the Monitoring Plan throughout on-study period. CRA's will also perform Investigational Product (IP) accountability at site.

A Site Close-Out visit (1 per site) will be conducted at each site following collection of all completed data and resolution of data queries.

17.4 AUDITING PROCEDURES

In addition to the monitoring visits outlined above, an investigational site may undergo a quality assurance audit. Linical Accelovance, Inc. or Client Company representatives or a regulatory agency such

as the FDA or EMEA may conduct the audit. If a regulatory agency requests an audit of the study site, the Investigator is required to inform Linical Accelovance, Inc. (or the Client Company) immediately.

17.5 DATA MANAGEMENT

The CRO will be responsible for the design and programming of the eCRF. A Data Management Plan will be developed describing all steps performed by Data Management. Data Management is responsible for creation of the Data Review Plan (containing both the checks programmed in the eCRF and manual checks to be performed), creation and execution of User Acceptance Test Plans prior to database go-live. Data cleaning of eCRF data entered by the sites and monitored by the CRAs will be performed by Data Management as described in the Data Management Plan, Data Review Plan and the CRO's applicable SOPs. All eCRF modifications are traced via an eCRF audit trail.

All data management activities will be performed using controlled and secure computing environments with validated software and systems (21 CFR Part 11 compliant) and well-designed workflow processes and procedures.

DM will be responsible for export of the dataset from the eCRF to submit to the statistical department for analysis.

17.6 QUALITY ASSURANCE

The study will be conducted according to the Sponsor and CRO Standard Operating Procedures. Quality Assurance (QA) will be performed under the responsibility of the Sponsor and CRO QA manager. They may periodically arrange inspections of the study by reviewing the data obtained as well as the procedural aspects. This may include on-site inspections and source data checks. Direct access to source documents is required for the purpose of these periodical inspections.

18 ETHICS AND REGULATORY REQUIREMENTS

18.1 ETHICAL CONDUCT OF STUDY

The study will be conducted in accordance with the ethical principles of the Declaration of Helsinki, the ICH-GCP Guidelines, the EU Clinical Trial Directive (2001/20/EG), and applicable regulatory requirements. The local investigator is responsible for the proper conduct of the study at the study site.

18.2 SUBJECT INFORMATION AND CONSENT

The investigator is responsible for ensuring that the patient fully understands the nature and purpose of the study. Information should be given in both oral and written form whenever possible. No patient should be obliged to participate in the study. Patients, their relatives, guardians or, if applicable, legal representatives must be given ample opportunity to enquire about details of the study. The information must make clear that refusal to participate or withdrawal from the study at any stage is without any prejudice to the patient's subsequent care. Patients must be allowed sufficient time to decide whether or not they wish to participate.

The patient must be made aware of (and give consent to) the fact that the monitors, auditors, the IEC and regulatory authorities will be granted direct access to the study patients source medical records without violating patient confidentiality, and to the extent permitted by applicable regulations. The patient should be informed that by signing the informed consent form the patient authorises such access.

The patient needs to give written informed consent before start of screening procedures. The patient will also be requested to give consent for the storage of tissue samples in the biobank.

The signed informed consent forms will be retained by the investigator and made available (for review only) to the study monitor, inspector and auditor upon request. The original signed informed consent form will be placed in the patients notes, a copy will be placed in the Regulatory Binder and the patient will be provided with a copy of the signed informed consent form.

18.3 REGULATORY APPROVAL

If legally required, the study will only start after having received written approval from the Regulatory (Competent) Authorities (CA). The regulatory application or submission for regulatory approval will be made by the sponsor as required by national law.

18.4 SUBJECT CONFIDENTIALITY

Individual patient medical information obtained as a result of this study is considered confidential and disclosure to third parties is prohibited. Such medical information may be given to the patient’s physician or to other appropriate medical personnel responsible for the patient’s well-being. The investigator will ensure that the patient’s anonymity will be preserved. In the eCRF or any other documents submitted to the Sponsor, the patients will not be identified by their names, but by an identification code, consisting of their initials and patient study number. Document not for submission to the Sponsor, i.e. the confidential patient identification code, original consent forms and source records will be maintained by the investigator in strict confidence. Source documents and original patient records may be inspected by countries’ regulatory health authorities therefore absolute confidentially cannot be guaranteed.

Data generated as a result of this study are to be available for inspection on request by the participating physicians, the sponsors' monitors, the IEC and the regulatory health authorities, including external site audits and inspections.

19 ADMINISTRATIVE MATTERS

19.1 INSURANCE OF SUBJECTS

The Sponsor has insured the risk by taking a liability insurance which is in accordance with the legal requirements in diverse countries. This insurance provides cover for damage to research patients through injury or death caused by the study.

The insurance applies to the damage that becomes apparent during the study. A condition is that the damage has been communicated in writing to the sponsor and the insurance company within this period.

19.2 USE OF INFORMATION AND PUBLICATION

Information concerning the study drug, patent applications, processes, unpublished scientific data, the IB and other pertinent information is confidential and remains the property of the Sponsor. Details should be disclosed only to the persons involved in the approval or conduct of the study. The investigator may use this information for the purpose of the study only. It is understood by the investigator that the Sponsor will use the information obtained during the clinical study in connection with the development of the drug and therefore may disclose it as required to other clinical investigators or to regulatory agencies. In order to allow for the use of the information derived from this clinical study, the investigator understands that he has an obligation to provide the Sponsor with all data obtained during the study.

The study will be considered for publication or presentation at (scientific) symposia and congresses. The investigator will be entitled to publish or disclose the results only after allowing the Sponsor to review all transcripts, texts of presentations and abstracts related to the study at least three months prior to the intended submission for publication or any other disclosure. This is necessary to prevent premature disclosure of trade secrets or patent-protected information and is in no way intended to restrict publication of facts or opinions formulated by the investigator. The Sponsor will normally inform the investigator of any objection or question arising within 30 days of receipt of the proposed publication material. After written approval is obtained, the manuscript is free for publication.

19.3 STUDY DOCUMENTATION AND DATA ARCHIVING

19.3.1 Investigator Site File

The investigator is responsible for maintaining adequate records to enable the conduct of the study to be fully documented and recorded. Copies of the study protocol, study approval letters, all original informed consent forms, a CD-ROM with all eCRFs, study medication dispensing and accountability logs, all correspondence pertaining to the study and any other documents relevant to the conduct of the study will be kept on file by the investigator for the maximum period of time required by local regulations. The default time period is fifteen (15) years or for at least 2 years after the granting of the last marketing authorisation in the EC (when there are no pending or contemplated marketing applications in the EC) or for at least 2 years after formal discontinuation of clinical development of the investigational product. The Sponsor will archive and retain all documents pertaining to the study for the lifetime of the product.

The investigator is responsible for maintaining a confidential patient identification code, which provides unique link between named source records and anonymous eCRF data for the sponsor. The investigator

must arrange for the retention of this confidential list for at least fifteen (15) years after the completion or discontinuation of the study.

No study document should be destroyed without prior written agreement between the investigators and the sponsor. Should the investigator elect to assign the study documents to another party, or move to another location, the sponsor must be notified.

For regulatory inspections, it will be necessary to have access to complete study subject records, provided that subject confidentiality is maintained.

19.4 PROTOCOL AMENDMENTS

Any changes to the study, which arise after approval of the protocol, must be documented as protocol amendments. A 'substantial' amendment is defined as an amendment to the terms of IEC application, or to the protocol or any other supporting documentation, that is likely to affect a significant degree:

- the safety or physical or mental integrity of the patients of the study;
- the scientific value of the study;
- the conduct or management of the study; or
- the quality or safety of any intervention use in the study

All substantial amendments will be submitted for approval to the IEC and to the CA as appropriate. Non-substantial amendments will be notified to the IEC and to the CA, as per IEC/CA procedures, and will be recorded and filed by the sponsor.

19.5 ANNUAL REPORTING

The Sponsor/investigator will submit a summary of the progress of the study to the IEC(s) once a year. Information will be provided on the date of inclusion of the first patient, numbers of patients included and numbers of patients that have completed the study, SAEs/ SARs, other problems, and amendments.

The Sponsor will submit, once a year throughout the clinical study, a safety report to the IEC and competent authorities of the concerned Member States. In addition to the expedited reporting of SUSARs, this safety report consists of:

- a list of all suspected (unexpected or expected) serious adverse reactions, along with an aggregated summary table of all reported serious adverse reactions, ordered by organ system, per study;
- a report concerning the safety of the patients, consisting of a complete safety analysis and an evaluation of the balance between the efficacy and the harmfulness of the medicine under investigation.

19.6 CLINICAL STUDY REPORT

The Sponsor will notify the IEC and the CA of the end of the study within a period of 90 days. The end of the study is defined as the last patient's last visit.

If the end of study is defined otherwise, this new definition should be given. In case the study is ended prematurely, the Sponsor will notify the IEC and the CA within 15 days, including the reasons for the premature termination.

Within one year after the end of the study, the Sponsor will submit a final clinical study report with the results of the study, including any publications/abstracts of the study, to the IECs and the CA.

20 REFERENCES

1. Dinmohamed AG, Visser O, van Norden Y, Blijlevens NM, Cornelissen JJ, Huls GA*, et al.* Treatment, trial participation and survival in adult acute myeloid leukemia: a population-based study in the Netherlands, 1989-2012. Leukemia **2016**;30:24-31.
2. Burnett A, Wetzler M, Lowenberg B. Therapeutic advances in acute myeloid leukemia. J Clin Oncol **2011**;29:487-94.
3. Terwijn M, van Putten WL, Kelder A, van der Velden VH, Brooimans RA, Pabst T*, et al.* High prognostic impact of flow cytometric minimal residual disease detection in acute myeloid leukemia: data from the HOVON/SAKK AML 42A study. J Clin Oncol **2013**;31:3889-97.
4. Dombret H, Gardin C. An update of current treatments for adult acute myeloid leukemia. Blood **2016**;127:53-61.
5. Alibhai SM, Leach M, Minden MD, Brandwein J. Outcomes and quality of care in acute myeloid leukemia over 40 years. Cancer **2009**;115:2903-11.
6. Palucka K, Banchereau J. Dendritic-cell-based therapeutic cancer vaccines. Immunity **2013**;39:38-48.
7. Barry M, Bleackley RC. Cytotoxic T lymphocytes: all roads lead to death. Nat Rev Immunol **2002**;2:401-9.
8. Sabado RL, Balan S, Bhardwaj N. Dendritic cell-based immunotherapy. Cell Res **2017**;27:74-95.
9. Schurch CM, Riether C, Ochsenbein AF. Dendritic cell-based immunotherapy for myeloid leukemias. Front Immunol **2013**;4:496.
10. Kalinski P, Edington H, Zeh HJ, Okada H, Butterfield LH, Kirkwood JM*, et al.* Dendritic cells in cancer immunotherapy: vaccines or autologous transplants? Immunol Res **2011**;50:235-47.
11. Kleindienst P, Brocker T. Endogenous dendritic cells are required for amplification of T cell responses induced by dendritic cell vaccines in vivo. J Immunol **2003**;170:2817-23.
12. Verdijk P, Aarntzen EH, Punt CJ, de Vries IJ, Figdor CG. Maximizing dendritic cell migration in cancer immunotherapy. Expert Opin Biol Ther **2008**;8:865-74.
13. Yewdall AW, Drutman SB, Jinwala F, Bahjat KS, Bhardwaj N. CD8+ T cell priming by dendritic cell vaccines requires antigen transfer to endogenous antigen presenting cells. PLoS One **2010**;5:e11144.
14. Mac Keon S, Ruiz MS, Gazzaniga S, Wainstok R. Dendritic cell-based vaccination in cancer: therapeutic implications emerging from murine models. Front Immunol **2015**;6:243.
15. de Gruijl TD, van den Eertwegh AJ, Pinedo HM, Scheper RJ. Whole-cell cancer vaccination: from autologous to allogeneic tumor- and dendritic cell-based vaccines. Cancer Immunol Immunother **2008**;57:1569-77.
16. Fox BA, Schendel DJ, Butterfield LH, Aamdal S, Allison JP, Ascierto PA*, et al.* Defining the critical hurdles in cancer immunotherapy. J Transl Med **2011**;9:214.
17. Ossenkoppele G, Schuurhuis GJ. MRD in AML: does it already guide therapy decision-making? Hematology Am Soc Hematol Educ Program **2016**;2016:356-65.
18. Dohner H, Estey E, Grimwade D, Amadori S, Appelbaum FR, Buchner T*, et al.*

Diagnosis and management of AML in adults: 2017 ELN recommendations from an international expert panel. Blood **2017**;129:424-47.

19. Janetzki S, Panageas KS, Ben-Porat L, Boyer J, Britten CM, Clay TM*, et al.* Results and harmonization guidelines from two large-scale international Elispot proficiency panels conducted by the Cancer Vaccine Consortium (CVC/SVI). Cancer Immunol Immunother **2008**;57:303-15.

21 APPENDIX A: ECOG/WHO PERFORMANCE STATUS

0 Normal activity
1 Symptoms, but nearly ambulatory
2 Some bed time, but to be in bed less than 50% of normal daytime
3 Needs to be in bed more than 50% of normal daytime
4 Unable to get out of bed
5 Death

Appendix B: CTC v4.0 criteria