Study Protocol

sdAb-based targeted radionuclide therapy of multiple myeloma: a feasibility study

UZBRU_ MUM

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Version history

Version	Date	Changes vs previous version
1.0	29/08/2017	Not applicable
2.0	30/03/2018	Two exclusion criteria removed Part 1 of the study (50 patients) split up in a prospective part (15 patients) and a retrospective analysis (35 patients). Two observations (demographics & medical history) moved to contact 2 instead of contact 1
2.1	30/01/2019	The inclusion/exclusion criteria of part 2 were modified: removal of exclusion criterion “patients treated for multiple myeloma in the past.

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Abbreviations

ASCT	Autologous stem cell transplantation
BM	Bone marrow
CR	Complete remission
CEA	Carcinoembryonic Antigen
CRF	Case Report Form
ELISA	Enzyme-Linked Immuno Sorbent Assay
EGFR	Epidermal growth factor receptor
Ga	Galium
GCP	Good Clinical Practice
GMP	Good Manufacturing Practice
ICH	The International Conference on Harmonisation
MM	Multiple myeloma
PET	Positron Emission Tomography
PSMA	Prostate Specific Membrane Antigen
SdAb	Single domain antibodies
SPECT	Single Photon Emission Computed Tomography
mAb	Monoclonal antibody
MFI	Mean Fluorescence Intensity
MMR	Macrophage Mannose Receptors
MRD	Minimal residual disease
TRNT	Targeted radionuclide therapy
HER2	Human epidermal growth factor receptor type 2
HPLC	High Performance Liquid Chromatography
kDa	Kilodalton
VIB	Vlaams Instituut voor Biotechnologie
VUB	Vrije Universiteit Brussel
ml	Milliliter

sdAb-based targeted radionuclide therapy of multiple myeloma: a feasibility study

Title:

"**UZBRU_MUM**" study

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CONFIDENTIALITY STATEMENT

The information provided in this document is strictly confidential and is available for review to investigators, potential investigators and appropriate Ethics Committees and authorities only. No disclosure should take place without the written authorization from VUB Technology Transfer, except to the extent necessary to obtain informed consent from potential patients.

1. STUDY ADMINISTRATIVE STRUCTURE

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Leading Ethical committee:

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2. INTRODUCTION

Multiple myeloma (MM) is an incurable cancer caused by malignant plasma cells in bone marrow (BM). Anti-myeloma drugs target either the myeloma cells directly or remodel the microenvironment of the tumor cell (1,2). By combining novel agents (e.g. proteasome inhibitors such as bortezomib or immunomodulatory drugs such as thalidomide and lenalidomide) with corticosteroids, followed by high dose chemotherapy and autologous stem cell transplantation (ASCT), high rates of survival and complete remission (CR) are currently achieved (3-4). CR is defined as the absence of monoclonal protein in serum and urine, less than 5% BM plasma cells, no increase of lytic bone lesions and disappearance of all pre-existing soft tissue plasmocytomas. However, disease recurrence is most often observed, due to the expansion of minimal residual disease (MRD) and the regrowth of chemo-resistant cancer cells in BM (5). Therefore, novel therapeutic strategies are warranted.
Due to their plasma cell origin, the hallmark of MM cells is the production of paraprotein (M-component), a monoclonal antibody (mAb) that is regularly secreted into the bloodstream. Upon diagnosis, the paraprotein is expressed on the surface of plasma cells in about 35% of MM patients (6-7). Each paraprotein sequence is unique for the patient's MM. It is defined by a unique antigen-binding site, which is frequently referred to as the 'idiotype'. Highly specific, anti-idiotype compounds might therefore be ideal tumor- and patient-specific vehicles for tumor imaging and therapy. Recently, Leung-Hagesteijn et al. reported a direct relation between bortezomib-resistance and the maturation stage of MM cells, whereby bortezomib-resistant malignant cells after MRD-regrowth are characterized by a more dedifferentiated phenotype and a concomitant expression of idiotype on their cell surface (8). This study provides further rationale to target MM cells in a MRD-setting with anti-idiotype vehicles since it provides a rescue strategy to eliminate cells that have otherwise become untreatable. However, the generation of truly anti-idiotypic vehicles is challenging because the few variable structures that give the M-component its unique nature are admixed within a large conserved framework.

Targeted radionuclide therapy (TRNT) is the technology in which peptides, mAbs or Ab fragments are coupled with therapeutic radionuclides. These vehicles interact with tumor-associated proteins that are expressed on the cancer cell surface. TRNT has become an attractive therapeutic application as it has the ability to target both the primary tumor site as well as disseminated disease. To date, only two agents are approved for commercial use, the radiolabeled anti-CD20 mAbs 90Y-ibritumomab and 131I-tositumomab to treat B cell non-Hodgkin lymphoma (blood-borne cancers). Due to the long blood half-life of mAbs, the systemic administration of radiolabeled mAbs is characterized by a prolonged presence of radioactivity in blood and highly perfused organs. Not surprisingly, myelotoxicity as a result of mAb-based TRNT is a well-known phenomenon and often a dose-limiting factor (9). Moreover, the dose delivered to carcinomas is often inadequate, due to limited penetration of mAb-based vehicles. Consequently, inadequate dose estimations often lead to inefficient treatment of

target lesions in clinical trials, limiting their success.
The integration of diagnostic testing (molecular imaging) for the presence of a molecular target is of interest to predict successful TRNT. This so-called “theranostic approach” aims to improve personalized treatment based on the molecular characteristics of cancer cells. Moreover, this strategy offers new insights in predicting the dose needed to treat and provides appropriate tools to monitor therapy responses. Due to their long circulation time, mAbs and most of their related fragments are not ideal for imaging, as it takes days before target-expressing lesions become visible.

Single domain antibodies or SdAb may solve some of the issues related to the use of mAbs for imaging and TRNT of cancer. SdAb are small (15 kDa) antigen-binding fragments derived from heavy-chain-antibodies of Camelidae. They are the smallest antibody-derived antigen-binding fragments and have superior characteristics compared to classical mAbs and their derived fragments for in vivo cell targeting (10). In the past, we have pioneered the successful evaluation of sdAb as in vivo diagnostic tracers (11). In terms of molecular imaging of cancer, sdAb have been directed to a variety of membrane-bound cancer cell biomarkers, such as CEA, EGFR, HER2, and PSMA. Because of their exceptional specificity of targeting, and the fact that they show to be functional after labeling with radionuclides, sdAb became valuable vehicles for nuclear imaging and TRNT (11,12). A first-in-human PET study with 68Ga-labeled anti-HER2 sdAb for breast cancer imaging was recently finalized in our university hospital (13,14), and a second PET study is planned for 2017, using an intratumoral macrophage-targeting anti-MMR sdAb.
More recently, we endeavored the use of sdAb as vehicles for TRNT in preclinical cancer models, through the therapeutic β--particle-emitting radionuclide Lutetium-177 (177Lu). These in vitro and in vivo proof-of-concept studies were very promising and showed strong signs of efficient and specific tumor treatment with absence of toxicity in non-target tissues (15-17). Crucial for the success of these therapy experiments were the measures that were taken to reduce renal retention of the radiolabeled sdAb, which could otherwise lead to kidney-related toxicities. This was partially successful by removal of the sdAb’ hexahistidine tag and the coinfusion with the plasma expander Gelofusin (17,18). For instance, we recently described the use of a 177Lu-labeled anti-HER2 sdAb for TRNT (17). SdAb-based targeted TRNT in mice bearing small subcutaneous HER2+ tumors led to a complete blockade of tumor growth and a significant difference in survival between the treated and the control. Similar exciting findings were recorded for a 177Lu-labeled sdAb to treat residual disease in a mouse model of MM (16). These preliminary data demonstrate for the very first time successful TRNT in preclinical cancer models using sdAb, and therefore suggest a potential of radiolabeled sdAb as a valuable adjuvant therapy candidate to treat residual and micrometastatic disease. In terms of TRNT, a low dose first-in-human biodistribution with therapeutic 131I-labeled anti-HER2 sdAb is planned for 2016 to assess safety, tolerability and biodistribution.

Our research group recently described, as proof-of-principle, the generation, characterization and use of anti-idiotypic sdAb as tools for specific targeting of MM cells in the 5T2MM murine

model (16). This model is reminiscent of the clinical situation where paraprotein is produced and expressed on the cancer cell membrane, CR is achieved after chemotherapy but the disease eventually recurs. After immunizing a dromedary with 5T2MM M-component/idiotype (5T2MMid) we identified high-affinity anti-idiotypic sdAb and demonstrated their anti-idiotypic nature and 5T2MM cancer-targeting properties in ELISA, BIAcore and FACS assays. Biodistribution studies and SPECT/micro-CT scans using ^{99m}Tc-labeled sdAb revealed specific in vivo targeting of the 5T2MMid. Treatment of 5T2MM mice in an MRD-like stage with ^{177}Lu-labeled sdAb resulted in significant signs of reduced tumor load. This study shows that anti-idiotypic sdAb are a new, sensitive, specific and non-invasive tool for imaging and therapeutic purposes and provides a rationale for their clinical evaluation as a personalized treatment option for MM patients expressing surface paraprotein.

3. STUDY OBJECTIVES

Part I
Primary objective of the clinical study:
Quantify the relative amount of patients that have expression of the paraprotein on the surface of multiple myeloma cells and determine a relevant threshold to select patients in part II of the study for anti-paraprotein sdAb generation

Part II
Primary objective of the clinical study:
Generate anti-paraprotein sdAb directed against the idiotype for 5 multiple myeloma patients, selected to have sufficient expression of paraprotein on the surface of their multiple myeloma cells

4. STUDY DESIGN

Single center, observational study that will be conducted prospectively for part I and II, and retrospectively for part III

5. INVESTIGATIONAL PLAN

5.1 Overall Study Design

Single center, observational

5.2 Study Groups

This study will be conducted in 2 parts, 50 patients in part I and 50 patients in part II, with a maximum of 100 patients for the entire study.

Part I will consist of:

- Part IA (15 patients): an additional laboratory analysis performed on excess material of bone marrow samples of multiple myeloma patients. The sampling will be performed in clinical routine and only excess materials (left over after samples for routine analysis are taken) will be used for study purposes. No additional sampling will be necessary for study purposes.
- Part IB (35 patients): a retrospective analysis of results of patients who underwent bone marrow biopsy between May 2009 and December 2017. Patients who were diagnosed with at least 10% malignant plasma cells and whose sample was analyzed for immunoglobulin phenotyping of both plasma cells and B lymphocytes will be included for further analysis.

Part II will consist of

- an additional laboratory analysis performed on excess material of bone marrow samples of multiple myeloma patients. The sampling will be performed in clinical routine and only excess materials (left over material after all samples for routine analysis are taken) will be used for study purposes. No additional bone marrow sampling will be necessary for study purposes.
- A blood sampling of maximally 10 ml by venous puncture (white serum tube), that will be used for study purposes

5.3 Time schedule

First patient: November 2017
Last patient: December 2031

5.3.1 Part IA

The individual patient schedule will be:

Contact 1:

- Informed consent
- Inclusion / exclusion criteria

Contact 2:

- Demographics
- Medical history

Bone marrow sampling performed in clinical routine /as standard of care
The clinical procedures will be performed as standard of care. There will be no additional study-related procedures.
The bone marrow sample will be analyzed as standard of care. Only leftover bone marrow material will be used for study purposes.
The following tests will be performed on leftover material: staining of membrane-bound paraprotein using anti-kappa and anti-lambda monoclonal antibodies for flow cytometry on non-permeabilized multiple myeloma cells (identified as positive using anti-CD38 staining) and analysis of binding using flow cytometry equipment.

Based on the results of part I, a threshold will be determined for the selection of patients in part II of the study.

Contact 1 and 2 can be performed at the same time, but the subject has to sign the informed consent prior to the performance of contact 2.

5.3.2 Part IB

This part will be conducted retrospectively. Informed consent by the patient is not required for this retrospective analysis.

5.3.3 Part II

The individual patient schedule will be:

Contact 1:

- Informed consent
- Inclusion / exclusion criteria
- Demographics
- Medical history

Contact 2: bone marrow sampling performed in clinical routine
There will be no additional study-related procedures at contact 2.
The bone marrow sample will be obtained and analyzed in clinical routine. Only left-over bone marrow material will be used for study purposes. Procedures will be identical to those in part I of the study.

After contact 2, the investigator will interpret the results of the bone marrow analysis for expression of the paraprotein on the membrane of multiple myeloma cells and compare the result with the threshold, to be determined after part I of the study. If the patient has a value above the threshold, the patient will be selected to continue the study.

Contact 3: A blood sampling of maximally 10 ml (white serum tube) will be obtained from the patient by puncture of a peripheral vein.

Subsequent contacts (if applicable): in case additional bone marrow samplings are performed as a standard of care procedure in the 10 years following inclusion, and in case there is leftover material (not needed for routinely performed tests) this leftover material could be used for study purposes.

The patient medical file will be checked on a regular basis to note any myeloma-related treatment, treatment response, time-to-relapse and time to next treatment.

5.4 Selection and withdrawal of subjects

5.4.1 Part IA

Inclusion Criteria:
Patients will only be included in the study if they meet all of the following criteria:

- Patients who have given informed consent
- Patients at least 18 years old
- Patient scheduled to undergo bone marrow sampling in clinical routine because of a clinically suspected or pathologically confirmed multiple myeloma.

Exclusion Criteria
Patients will not be included in the study if one of the following criteria applies:

- Patients who are unwilling and/or unable to give informed consent

Withdrawal of patients
The patient may withdraw from the study at any time without giving reasons and without any disadvantageous consequences for his/her subsequent medical care.

The investigator may withdraw a patient from the study at any time in case of intercurrent illness, adverse event, therapeutic procedure or as a general rule if further participation in the study is believed to be to the patient's detriment.

Any patient withdrawn from the study before the scheduled completion of the study procedures will be considered an early withdrawal, regardless of the reason for withdrawal.

Patients withdrawn from the study will be replaced.

A patient participating in part I of the study has completed his study participation at the end of contact 2.

5.4.1 Part IB

Inclusion Criteria:

Patients who underwent bone marrow biopsy between May 2009 and December 2017 and:

- who were diagnosed with at least 10% malignant plasma cells in the bone marrow sample
- whose sample was analyzed for immunoglobulin phenotyping of both plasma cells and B lymphocytes

Exclusion Criteria
Patients will not be included in the study if no immunofixation analysis was performed on blood samples

5.4.2 Part II

Inclusion Criteria:
Patients will only be included in the study if they meet all of the following criteria:
- Patients who have given informed consent
- Patients at least 18 years old
- Patients scheduled to undergo bone marrow sampling in clinical routine because of a clinically suspected or pathologically confirmed multiple myeloma.

Exclusion Criteria
Patients will not be included in the study if one of the following criteria applies:
- Patients who cannot communicate reliably with the investigator
- Patients who are unlikely to cooperate with the requirements of the study
- Patients who are unwilling and/or unable to give informed consent
- Patients at increased risk of death from a pre-existing concurrent illness
- Patients who participated already in part I of this study

Withdrawal of patients
The patient may withdraw from the study at any time without giving reasons and without any disadvantageous consequences for his/her subsequent medical care.

The investigator may withdraw a patient from the study at any time in case of intercurrent illness, adverse event, therapeutic procedure or as a general rule if further participation in the study is believed to be to the patient's detriment.

Any patient withdrawn from the study before the scheduled completion of the study procedures will be considered an early withdrawal, regardless of the reason for withdrawal.

Patients withdrawn from the study will be replaced.

A patient participating in part II of the study has completed his study participation 10 years after inclusion (additional per-protocol analysis on left-over bone marrow sample are allowed until 10 years after inclusion).

5.5 Clinical Laboratories, Technical Department(s), Institution(s) Providing Clinical Study Services

The local department of clinical chemistry and hematology will perform the study related laboratory testing of bone marrow samples for part IA and part II of the study. Also the retrospective analysis of part IB will be performed by this department. For part IA, an additional immunoglobulin phenotyping analysis on left-over bone marrow sample will be performed using both cytoplasmatic and membrane staining using flow cytometry. For part IB,

retrospectively results of patients who underwent bone marrow biopsy with such flow cytometry analysis will be reanalyzed to determine membrane expression of the paraprotein. This will be possible if a relatively high amount of malignant plasma cells is present in the sample (10% as a threshold is used). In that case, we will assess if these plasma cells can be distinguished on the scatter plots because of their typical forward and side scatter characteristics. If this population is overlapping with other cell populations on flow cytometry, the sample will be excluded for further analysis. Mean fluorescence intensity (MFI) of membrane-bound immunoglobulins (IgM, IgD, IgG, IgA, kappa and lambda) on the plasma cells will be quantified using isotype control staining as a reference. Also the cytoplasmatic expression of immunoglobulins on plasma cells will be quantified on the available flow cytometry data. The obtained data will be correlated to the blood-based immunofixation assay that was performed in the selected patients. This allows to identify the type of immunoglobulin that was secreted in the blood of the patient, and to distinguish irrelevant fluorescent signal produced by non-secreted immunoglobulins from the relevant signal produced by membrane-bound paraprotein immunoglobulin. Because this method can only be performed in patients who had a high number of malignant plasma cells in their sample, and since interference of other cells cannot be entirely excluded using this method, we will further confirm these results using the prospectively collected bone marrow samples of part IA of this study.

The In vivo cellular and molecular imaging laboratory of the VUB will process the blood sample drawn via venous puncture of the patient. The patient-specific M-component will be purified through chromatography procedures. Next, sdAbs will be generated against the purified patient-specific paraprotein in collaboration with the VIB Nanobody Core located at the VUB. In short, a sdAb-library is generated after immunization of a dromedary with the paraprotein. Peripheral blood lymphocytes are collected, followed by an amplification of the RNA encoding for the variable parts of the specific heavy-chain-only antibodies (= single-domain antibodies, sdAbs). These sequences are ligated into phagemid vectors, introduced into bacterial cells and the resulting sdAb library is phage-displayed. Paraprotein-targeting sdAbs are obtained via biopannings of the sdAb-phage library on immobilized paraprotein in the presence of normal human serum and sequencing of the obtained clones.

The anti-idiotypic nature and affinity towards paraprotein, and the stability in human serum of the selected sdAbs are then characterized, using techniques like ELISA, BIAcore and HPLC. Gamma-HPLC profiles of ^{99m}Tc-labeled sdAbs added to normal human serum that is spiked with increasing concentrations of paraprotein will provide surrogate measurements of specificity and affinity for the patient's idiotype. The *in vivo* behavior of the different sdAbs will be evaluated in animal models. Binding of sdAbs to the membrane of patient-derived malignant plasma cells (cytospin of BM aspirates) will be monitored via immunohistochemistry or flow cytometry if such cells are available as left-over material of routinely performed bone marrow sampling. Based on these results, a lead sdAb will be selected.

The selected lead sdAb from will be radiolabeled with the therapeutic radionuclide ^{177}Lu using

a functionalized bifunctional chelator through previously established procedures, but now translated into a GMP-procedure together with the Nuclear Medicine department of the UZ Brussel. After radiolabeling, binding on immobilized paraprotein will be evaluated through radioligand binding studies, as well as stability in human serum. Binding towards paraprotein in patient blood samples at different disease stages will be analyzed through HPLC.
At the end of this experimental part, we aim to deliver a pure and functional radiolabeled patient-specific anti-idiotype sdAb, which is completely characterized and will be ready for personalized therapy. The obtained HPLC results, circulating paraprotein levels, membrane paraprotein-positivity and other clinical parameters (including BM Giemsa stainings) will provide us with detailed information on the fraction of patients that would be eligible for sdAb-based TRNT and the time-frame in which they should ideally be treated in a future clinical trial.

5.6 Potential risks and benefits to human subjects

Potential Risks

The bone marrow sampling is performed in clinical routine and does not induce additional risks as compared to patient care outside of this study.

The puncture of a vein for blood sampling, performed in patients participating in part II of the study, is performed for study purposes. This procedure might (rarely) cause pain, bleeding, bruising or infection localized around the injection site. Similarly, some subjects might feel dizzy or even faint during the procedure. The clinical team that will perform this procedure will do all they can to keep these discomforts to a minimum.

Benefits

For the cancer patients participating in part II of the study, there is the potential that a sdAb against their paraprotein is developed. It is not the primary goal of the study to use this sdAb to treat the patient. If however available and if ethically and clinically acceptable, it might be considered as an individualized therapy in a future clinical trial.

The information obtained thanks to this study can contribute to the development of a new therapeutic for the patient-specific treatment of multiple myeloma patients.

5.7 Outcome measures of safety, tolerability and tumor targeting

5.7.1 Primary outcome measures for part I

Using flow cytometry analysis on leftover bone marrow samples, the following outcome measures will be reported:

- Amount of patients that shows membrane expression of the paraprotein (or part of the paraprotein) on a subset or all of their multiple myeloma cells in the bone marrow sample
- Fraction of multiple myeloma cells within a the sample of one patient that has membrane expression of the paraprotein
- Variation in intensity (expression level) of paraprotein expression in such membrane-expressing multiple myeloma cells

Based on these observations, a cutoff value for sufficient expression of paraprotein on the membrane of multiple myeloma cells will be determined.

5.7.2 Primary outcome measures for part II

- Fraction of patients (n=5) for whom a sdAb could be generated that binds to the idiotype of the paraprotein
- Amount of paraprotein-targeting sdAbs generated per patient and amount of sdAb families identified
- Affinities of generated paraprotein-targeting sdAbs using BIAcore technology
- Affinities of generated paraprotein-targeting sdAbs using ELISA techniques

5.7.3 Secondary outcome measure for part II

- Binding of paraprotein-targeting sdAb to membrane-expressed paraprotein of multiple myeloma cells, obtained as leftover material from routinely performed bone marrow sampling in the 10 years after inclusion of the study subject

5.8 Statistical methods planned

5.8.1 Sample size justification

This study is of exploratory nature and is intended to generate a proof-of-concept that paraprotein-targeting sdAbs can be generated for paraprotein derived from human subjects. The sample size was chosen based on the available budget for nanobody generation

5.8.2 Paraprotein-targeting evaluation

Classical descriptive statistics for categorical variables (N, n and %) will be used. Obtained sdAbs will be evaluated in normal mice for normal biodistribution and results will be analyzed using two-way Anova.

6. DATA HANDLING AND RECORD KEEPING

A paper CRF will be completed for each screened subject. This CRF will include specific pages for inclusion and exclusion criteria conclusions, for reporting each visit and the reporting of biological results. The investigator will review, approve and validate each completed CRF; the investigator's signature (validation) serving as attestation of the investigator's responsibility for ensuring that all clinical and laboratory data entered on the CRF are complete, accurate and authentic.

7. DIRECT ACCESS TO SOURCES

7.1 Source data and documents

Source data are all the information in original records and certified copies of original records of clinical findings, observations, or other activities in the study, which are necessary for the reconstruction and evaluation of the study. Source data are contained in source documents (originals or certified copies).

All information recorded on the CRFs for this study must be consistent with the subject's source documentation. The source documents (i.e. hospital records) must contain the first, middle (where applicable) and the last name of the subject, the date of birth, the information that the informed consent was signed and dated prior to the study, the date of entry in the study, the date of the visits, the code number of the subject, and the date of completion of the study.

Source records should be preserved for the maximum period of time permitted by local requirements (30 years in Belgium). For each subject enrolled, the investigator will indicate in the source record(s) that the subject participates in this study, and record the following information: subject number, identification of the subjects (name, address, etc.), and main reason for discontinuation, if applicable.

7.2 Direct access to source data and documents

Secret must be respected regarding the identity of persons participating in a clinical study and all information of a personal or medical nature concerning these subjects. Any information which might permit identification of subject must be kept confidential. This confidentiality must be guaranteed to all subjects participating in the study.

In all reports involving the subjects included in the study, each subject will be identified only by a subject number with four characters and the birthday year. The investigator will ensure that the confidentiality of subjects' data will be preserved. On CRFs or any other documents, the subjects will not be identified by their names, but by an identification system, which consists of their number in the study. Documents that identify the names of participants against their study number, i.e. the confidential subject identification log will be maintained by the investigator in strict confidence.

The investigator will be personally responsible for the updating of a subject identification log which relates subject numbers to the names and means of contact of each subject in the study. This log will be kept in the investigator's file so that the investigator can contact the subjects. The confidentiality of these data will be preserved in accordance with the applicable national and/or local laws and regulations on personal data protection.

Monitors, auditors and other authorized agents will be granted direct access to study subject's original medical records for verification of clinical study procedures and/or data, without violating the confidentiality of the subjects, to the extent permitted by the law and regulations. In any presentations of the results of this study at meetings or in publications, the subjects'

identity will remain confidential.

8. QUALITY CONTROL

The investigator will permit direct access of the study monitors and appropriate regulatory authorities to the study data and to the corresponding source data and documents to verify the accuracy of this data.

The investigator agrees to allow the monitor access to any or all of the study materials needed for the monitor to properly review the study progress. The investigator (or deputy) agrees to assist the monitor in resolving any problem that may be detected during the monitoring visit. Monitor should write a monitoring report of visit performed on investigator sites and a monitoring letter to the visited investigator.

8.1 Audits and inspections

The investigator should understand that source documents for this study should be made available to appropriately qualified personnel from the Sponsor, Quality Assurance Unit or its designees, or to Health Authority Inspectors after appropriate notification. The investigator's site, facilities and all data (including source data) and documentation could be made available for audit by an independent quality assurance unit and for EC or regulatory authorities according to the guideline of the international conference on harmonization (ICH)-good clinical practice (GCP). The investigator agrees to co-operate with the auditor during his/her visit and will be available to supply the auditor with CRFs or other files necessary to conduct that audit. Any findings will be strictly confidential. The main purposes of an audit or inspection are to confirm that the rights and well-being of the subjects have been adequately protected, and that all data relevant for the evaluation of the investigational product have been processed and reported in compliance with GCP and applicable regulatory requirements. The verification of the CRF data must be done by direct control of source documents.

9. ETHICS

9.1 Ethics Committee Approval

The investigator will obtain, from the Ethics Committee of UZ Brussel a prospective approval of the clinical study protocol and corresponding informed consent form(s); modifications to the protocol and corresponding informed consent form(s), and advertisements (i.e., directed at potential research subjects) for study recruitment.

The Ethics Committee of UZ Brussel operates in compliance with European regulations and in conformance with applicable ICH-GCP guideline.

In the event that the Ethics Committee requires, as a condition of approval, substantial

changes to the clinical protocol, or in the event of a decision to modify the previously accepted clinical protocol, the investigator will submit (i.e., in advance of implementing the change) a Protocol Amendment describing any change to the clinical protocol that significantly affects the safety of the subjects. For changes that do not affect critical safety assessments, the revisions to the clinical protocol will be notified to the Ethics Committee(s).

9.2 Ethical and scientific conduct of the clinical study

The clinical study will be conducted in accordance with the current Ethics committee-approved clinical study protocol; Declaration of Helsinki principles; ICH Guidelines on GCP; and relevant policies, requirements, and regulations.

10. FINANCING AND INSURANCES

10.1 Financing

The study is financed by Vrije Univeristeit Brussel, through a grant provided by stichting tegen kanker

10.2 Insurances

The subjects taking part in this study will be covered by the insurance taken by the UZ Brussel, if they were to suffer any prejudice as a result of taking part in the study and according to ICH-GCP requirements, the institution has taken out personal liability insurance with an Insurance company following the Belgian regulations. This insurance was taken out with Ethias NV.

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