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<b>Study Protocol</b>		

<b>Study Protocol</b>	
<b>Protocol Number</b>	AAA-Annexin-03 / NCT03232580
<b>Investigational Product</b>	Kit for the preparation of <sup>99m</sup> Tc-rhAnnexin V-128
<b>Active substance</b>	rhAnnexin V-128
<b>Radiolabeled Imaging Product</b>	<sup>99m</sup> Tc-rhAnnexin V-128
<b>Trial Phase</b>	Proof of Concept and Phase II
<b>Trial Title</b>	Phase II Study of <sup>99m</sup> Tc-rhAnnexin V-128 Radionuclide Imaging in Patients with Clinical Suspicion or Confirmed Diagnosis of Spondyloarthritis (SpA)
<b>Short Trial Title</b>	<sup>99m</sup> Tc-rhAnnexin V-128 in patients with SpA
<b>Version and Date</b>	v4.0 – 13 March 2018
<b>Investigators</b>	<b>Principal Investigator:</b> Dr [REDACTED], MD  <b>Co-Investigators:</b> Dr [REDACTED], MD
<b>Trial Sponsor</b>	Advanced Accelerator Applications USA Inc.
<p>The concepts and information contained herein or generated during the study are considered proprietary and shall not be disclosed in whole or in part without the expressed written consent of Advanced Accelerator Applications USA Inc.</p>	
<p>This study is to be completed according to the guidelines of Good Clinical Practice (GCP) and conducted in full compliance with the World Medical Association Declaration of Helsinki and its most recent amendments.</p>	

## SPONSOR SIGNATORY APPROVAL PAGE

Key Personnel - Advanced Accelerator Applications		
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**Study Protocol****INVESTIGATOR ENDORSEMENT PAGE**

I agree to conduct the study as outlined in the protocol entitled “Phase II Study of <sup>99m</sup>Tc-rhAnnexin V-128 Radionuclide Imaging in Patients with Clinical Suspicion or Confirmed Diagnosis of Spondyloarthritis (SpA)” in accordance with the guidelines and all applicable government regulations. These guidelines and regulations include, but are not limited to:

1. Permission to allow the Sponsor and the regulatory agencies to inspect study facilities and pertinent records at reasonable times and in a reasonable manner that ensures subject confidentiality. If this study is to be inspected by a regulatory agency, the Sponsor should be notified as soon as possible;
2. Submission of the proposed clinical investigation, including the protocol and the consent form to a duly constituted Institutional Review Board (IRB) or Ethics Committee (EC) for approval and acquisition of written approval for each, prior to the use of the test article;
3. Use of written informed consent that is obtained prior to administration of test article or any non-routine procedures that involve risk, and that contains all the elements of consent as specified in the federal regulations and has been previously approved by the Sponsor and the IRB/EC;
4. Submission of any proposed change in or deviation from the protocol to the IRB/EC using a signed formal amendment document approved by the Sponsor. Any proposed changes or deviations from the protocol require that the informed consent also reflect such changes or deviations and that the revised informed consent be approved by the IRB/EC;
5. Documentation and explanation of individual protocol deviations on the appropriate case report form page or in letters to the Sponsor;
6. Reports of serious adverse events to the Sponsor/CRO within 24 hours by telephone and a written report of the serious adverse event within 72 hours after the Investigator’s initial receipt of the information;
7. Reporting of Serious Adverse Events (SAEs) according to ICH/GCP and Regulatory Standards. SAEs will be reported from the signing of the informed consent and followed until resolution or determined to be not clinically significant;
8. Submission of timely progress reports to the IRB/EC and Sponsor at appropriate intervals on a schedule determined by the IRB/EC.

Regulations require an Investigator to prepare and maintain adequate and accurate case histories designed to record all observations and other data (such as test article accountability) pertinent to the investigation on each individual enrolled in the study. These records must be maintained by the Investigator for a period of 2 years or a period of time determined by the Sponsor following the date a marketing application is approved for the indication for which it is being investigated, or, if no application is to be filed or if the application is not approved for such indication, until 2 years or a period of time determined by the Sponsor after the investigation is discontinued and the appropriate regulatory authorities are notified.

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In addition, I agree to provide all the information requested in the case report form in a manner to assure legibility and accuracy. To this end, I shall carefully follow the instructions for completing case report forms.

I also agree that all information provided to me by the Sponsor, including protocols, case report forms, and verbal and written information, will be kept strictly confidential and confined to the clinical personnel involved in conducting the study. It is recognized that this information may be related in confidence to the IRB/EC. I also understand that reports of information about the study or its progress will not be provided to anyone who is not involved in the study other than to the Principal Investigator, or in confidence to the IRB/EC or to the legally constituted regulatory authorities.

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Principal Investigator Signature

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Date of Signature

## 1 SYNOPSIS

<b>Investigational Medicinal Product</b>	Kit for the Preparation of Tc-99m Recombinant Human Annexin V-128 for Injection
<b>Title of the study</b>	Phase II Study of <sup>99m</sup> Tc-rhAnnexin V-128 Radionuclide Imaging in Patients with Clinical Suspicion or Confirmed Diagnosis of Spondyloarthritis (SpA).
<b>Clinical Center</b>	Cedars-Sinai Medical Center 8700 Beverly Blvd., 1258 Los Angeles, CA 90048 United States
<b>Investigators</b>	Dr [REDACTED], MD Dr [REDACTED], MD
<b>Corporate Sponsor</b>	Advanced Accelerator Applications USA, Inc.
<b>Subject's Indication</b>	Patients with Clinical Suspicion or Confirmed Diagnosis of Spondyloarthritis (SpA)
<b>Objectives</b>	<p>Primary Objective</p> <ul style="list-style-type: none"> <li>- To determine the magnitude and dynamic range of <sup>99m</sup>Tc-rhAnnexin V-128 uptake in disease affected sacro-iliac or lumbar spine joints, in patients with clinical suspicion or confirmed diagnosis of Spondyloarthritis (SpA).</li> </ul> <p>Secondary Objectives:</p> <ul style="list-style-type: none"> <li>- To determine the clinical utility of <sup>99m</sup>Tc-rhAnnexin V-128 SPECT/CT imaging in the identification of chronic osteoarthritic active sites of SpA compared with conventional imaging (Magnetic Resonance Imaging), in patients with disease affected sacro-iliac, or lumbar spine joints.</li> <li>- To assess the presence of antibodies against rhAnnexin V-128 at baseline and post-treatment</li> <li>- To determine the localization pattern and magnitude of focal uptake of <sup>99m</sup>Tc-rhAnnexin V-128 within the abdomen in patients with clinical suspicion or confirmed diagnosis of Spondyloarthritis (SpA).</li> </ul> <div style="background-color: black; height: 80px; width: 100%; margin-top: 10px;"></div>
<b>Rationale</b>	Annexin V is an endogenous human protein that binds to phosphatidylserine (PS), a constitutive anionic phospholipid of the plasma cell membranes in all

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	mammalian cells, that is expressed on the surface of physiologically stressed, depolarized, or apoptotic cells only. Technetium-99m ( <sup>99m</sup> Tc) is a medical isotope widely used in diagnostic imaging. <sup>99m</sup> Tc-labeled annexin V-128 has demonstrated the ability to image cellular stress, apoptosis, and necrosis in a variety of diseases with strong inflammatory component including rheumatoid arthritis, prosthetic joint infection, myocarditis, acute heart transplant rejection, or Crohn's disease in both, animal models and in humans.
<b>Planned number of patients</b>	Twenty evaluable patients with clinical suspicion or confirmed diagnosis of spondyloarthritis due to ankylosing spondylitis, or associated with other known clinical conditions like inflammatory bowel disease, psoriatic arthritis, or undifferentiated spondyloarthropathy. Five patients will be recruited in the first part of the trial, as a Proof of Concept (PoC) phase. The PoC phase will assess the imaging potential of <sup>99m</sup> Tc-rhAnnexin V-128 in terms of imaging quality, disease-lesion radiotracer uptake and medical relevance to enable the decision-making to terminate or to continue the clinical investigation completing the enrollment with the remaining 15 patients.
<b>Study design and Methodology</b>	This is a single-centre, open label, PoC, Phase II study. Patients who have signed the informed consent and are eligible to participate in the study will undergo the following assessments: <ul style="list-style-type: none"> <li>• At screening visit, eligible and consented patients who have clinical suspicion or confirmed diagnosis of SpA will undergo a physical examination, vital signs (systolic and diastolic blood pressure, heart rate), blood analysis [CBC with automated differential, general chemistry panel (SMA-20) including CRP, PT/PTT, INR (international normalized ratio for PT values), assessment of anti-annexin V-128 antibodies] and urinalysis. In addition, disease activity score assessments (BASDAI, BASFI, BASMI and MASES) will also be performed. Screening laboratory assays, physical examination and vital signs will be performed 24 hours to 14 days before <sup>99m</sup>Tc-rhAnnexin V-128 imaging. Within 14 days of <sup>99m</sup>Tc-rhAnnexin V-128 scanning patients will undergo T1 and STIR without contrast Magnetic Resonance Imaging (MRI) of the sacro-iliac and lumbar spine joints.</li> <li>• At Day 0 (the day of intravenous injection and imaging with <sup>99m</sup>Tc-rhAnnexin V-128), a dedicated lumbosacral spine and abdomen SPECT/CT starting 60 minutes after injection of tracer followed by a whole body planar scan and spot views on selected areas will be performed. The same SPECT/CT and whole body planar scans will be repeated at 2 hrs after injection of tracer except the spot views unless they suggest to be repeated. Vital signs assessment will also start 15 minutes before and for 2 hours after injection of radiotracer.</li> <li>• At Day 1 (one day after the administration of <sup>99m</sup>Tc-rhAnnexin V-128), 15\20 patients will be contacted over the phone for assessment of possible adverse events (e.g. allergic reaction such as rash, edema). 5</li> </ul>

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	<p>patients will return for an optional whole body planar scan at 24hrs post injection as well as spot views, if deemed appropriate by the investigator. Safety information will be collected at this time for these 5 patients.</p> <ul style="list-style-type: none"> <li>• At Day 30 <math>\pm</math> 3 days, all patients will return to the clinic for a physical examination and a final blood sampling to rule out the development of anti-annexin V-128 antibodies and for the following blood analysis and urinalysis: CBC with automated differential, general chemistry panel (SMA-20) including CRP, PT/PTT, INR (international normalized ratio for PT values).</li> <li>• Once the first 5 patients have been administered with <math>^{99m}\text{Tc}</math>-rhAnnexin V-128 and have performed the imaging at Day 0, a Data Monitoring Committee (DMC) will review the images. The visual assessment of the scans in these first 5 patients may support the continuation or the termination of the Phase II study. In the first case, the enrollment of 15 additional patients will be required.</li> </ul>
<b>Treatment</b>	The treatment will consist of $^{99m}\text{Tc}$ -rhAnnexin V-128. Following the reconstitution and labelling of rhAnnexin V-128, the imaging product will be administered as a single intravenous bolus of 350 MBq $\pm$ 10% over 10-20 seconds at Day 0 for the purpose of SPECT/CT imaging.
<b>Inclusion/ exclusion criteria</b>	<p><b>Inclusion criteria</b></p> <p><b>For the first 5 patients enrolled in the POC part:</b></p> <ol style="list-style-type: none"> <li>1. Patients with clinical suspicion or confirmed diagnosis of SpA, based on the ASA criteria with active symptoms including back, hip or buttock pain prior to: <ul style="list-style-type: none"> <li>• A change in NSAID therapy or</li> <li>• A change in non-biologic DMARD or</li> <li>• A start of non-biologic DMARD.</li> </ul> </li> </ol> <p><b>For the next 15 patients enrolled in the Phase II part:</b></p> <ol style="list-style-type: none"> <li>1. Patients with clinical suspicion or confirmed diagnosis of SpA, based on the ASAS criteria with active symptoms including back, hip or buttock pain prior to: <ul style="list-style-type: none"> <li>• A change in NSAID therapy or</li> <li>• A change in non-biologic DMARD or</li> <li>• A start of non-biologic DMARD or</li> <li>• A start of biologic DMARD</li> </ul> </li> </ol> <p><b>For all patients:</b></p> <ol style="list-style-type: none"> <li>2. Age over 18 years old.</li> <li>3. Signed Informed Consent Form.</li> </ol>

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	<p><b>Exclusion criteria</b></p> <ol style="list-style-type: none"> <li>1. Pregnancy or lactation</li> <li>2. Liver impairment (ALT, AST or Bilirubin &gt; 2 ULN) at screening visit or baseline.</li> <li>3. Kidney impairment (serum creatinine &gt; 1.5 mg/dL)</li> <li>4. History of any disease or relevant physical or psychiatric condition or abnormal physical finding which may interfere with the study objectives at the investigator judgment.</li> <li>5. Know hypersensitivity to the investigational product or any of its components.</li> <li>6. Contraindication(s) to the MRI procedure (claustrophobia, prosthetic valve, pacemaker, inability to lie still in a supine position)</li> <li>7. Participation in another clinical trial within 4 weeks before study inclusion.</li> </ol>
<p><b>Study duration and assessments:</b></p>	<p>Maximum 7 weeks including the screening period, for an individual patient.</p>
<p><b>Safety</b></p>	<p>Venous blood samples will be drawn for each patient at screening and at Day 30 for laboratory analysis (CBC with diff., SMA-20 including CRP, PT/PTT, INR).</p> <p>To rule out the development of anti-rhAnnexin V-128 antibodies, venous blood samples will also be drawn for each patient at screening and Day 30. Assays for anti-rhAnnexin V-128 IgG and IgM antibodies will be performed in serum samples by ELISA. For this purpose, 10 mL of blood will be collected at screening visit and at Day 30.</p> <p>24 hours after injection, at Day 1, each patient will be called to check if they experienced any Adverse Events.</p> <p>All patients will undergo a physical examination at screening and at Day 30. Vital signs (systolic and diastolic blood pressure, heart rate) will be performed by all patients at screening and Day 0 (before and after injection of the radiotracer). If there is any Adverse Event(s), they will be reported and recorded in the Electronic Case Report Form (eCRF).</p>
<p><b>Statistics</b></p>	<p>The number of patients in this study is not based on statistical power considerations. Five patients will be enrolled in the PoC phase, and 15 additional patients in the second part of the study, which is believed to provide sufficient data to demonstrate the potential of <sup>99m</sup>Tc-rhAnnexin V-128 imaging in the assessment of disease severity in SpA patients and plan for future clinical studies.</p> <p>Adverse events will be listed on an individual basis, including relationship, and severity, and will be summarized by System Organ Class (SOC) and Preferred Term (PT). Patients with more than one adverse event within a particular SOC</p>

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and PT will be counted only once for that SOC and PT. The incidence of adverse events will also be summarized by severity.

All other safety variables will be tabulated at each measuring time together with descriptive statistics, if appropriate. Hematology, clinical chemistry, urinalysis and immunogenicity data will be analyzed. Similarly, descriptive statistics will also be provided for vital signs, physical examination, etc.

Efficacy by  $^{99m}\text{Tc}$ -rhAnnexin V-128 uptake will be assessed qualitatively and semi-quantitatively in sacro-iliac joints and in the lumbar spine of patients with clinical suspicion or confirmed diagnosis of SpA. Sensitivity and specificity of  $^{99m}\text{Tc}$ -rhAnnexin V-128 SPECT/CT imaging compared to MRI will be evaluated based on the clinical follow-up information as the standard of truth for disease diagnosis.

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Visit schedule – Table 1

Study Procedures	Screening (Within 14 days prior to Day 0)	Day 0	Day 1 24 hours <sup>1</sup>	Day 30
Written informed consent	x			
Inclusion/exclusion criteria	x			
Medical history	x			
Concomitant medications	x	x	X	X
Disease assessment (BASDAI, BASFI, BASMI, MASES)	x			
Physical examination	x			x
Vital signs (BP, HR)	x	x		
Lab analysis (hematology, PT/PTT/INR biochemistry; urine)	x			x
Immunogenicity by ELISA <sup>2</sup>	x			x
Pregnancy test	x	x		
MRI (within 14 Days of rh-Annexin V-128 scanning)	x <sup>3</sup>			
rh-Annexin V-128 administration and scan (SPECT/CT and planar imaging)		X <sup>4</sup>		
Whole Body Planar Imaging + spots			X <sup>4</sup>	
High Resolution spot images of hands, feet, any symptomatic joint group		X <sup>4</sup>	X <sup>4</sup>	
Adverse Events	x	x	X	x

<sup>1</sup> 24 hours after injection of <sup>99m</sup>Tc-rh-Annexin V-128, each patient will be called over the phone to assess if they experienced any adverse events such as allergic reactions (edema, rash).

<sup>2</sup> Anti-rhAnnexin V-128 IgG and IgM antibodies will be quantified in serum samples by ELISA. For this purpose 10 mL blood samples will be collected at screening and Day 30 post administration of test agent. Serum will be prepared, divided into six aliquots and frozen (-80°C). Three out of six serum aliquots will then be shipped to the central laboratory at [REDACTED]. The 3 remaining aliquots will be sent to [REDACTED] with an additional shipment.

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<sup>3</sup> A whole lumbar spine MRI must be performed prior to the enrolment date, or within 14 days of <sup>99m</sup>Tc-rh-Annexin V-128 scanning prior to changing NSAID therapy or non-biologic DMARD or starting non-biologic DMARD for the first 5 patients and prior to changing NSAID therapy or non-biologic DMARD or starting non-biologic DMARD or biologic DMARD for the next 15 patients.

<sup>4</sup> First five patients after which the process will be re-evaluated:

- All patients will have whole body planar imaging at 60 minutes post injection.
- At two hours post injection do a whole body planar scan and SPECT/CT with onboard isotope with no spot views unless initial views suggest otherwise. The whole body planar scan adds 15-20 minutes; each spot adds 5 minutes
- The SPECT/CT and spot imaging will be done for first 5 patients after which the protocol will be re-adjusted.
- An optional whole body planar scan will be done at 24hrs post injection for 5 patients as well as spot views, if deemed appropriate by the investigator.

## 2 LIST OF ABBREVIATIONS

AAA	Advanced Accelerator Applications
AE	Adverse Event
ACR	American College of Rheumatology
AS	Ankylosing Spondylitis
ASAS	Assessment of Spondylo-Arthritis Society
BASDAI	Bath Ankylosing Spondylitis Disease Activity Index
BASFI	Bath Ankylosing Spondylitis Functional Index
BASMI	Bath Ankylosing Spondylitis Metrology Index
Bi-DMARD	Biological Disease Modifying Anti-Rheumatic Drug
BUN	Blood Urea Nitrogen
CIA	Collagen Induced Arthritis
CRF	Case Report Form
CRO	Clinical Research Organization
CRP	C-Reactive Protein
CT	Computed Tomography
DAS28	Disease Activity Score
DMARD	Disease Modifying Anti-Rheumatic Drug
DMC	Data Monitoring Committee
ELISA	Enzyme-Linked Immuno-absorbent Assay
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GP	General Practitioner
Hb	Hemoglobin
HPLC	High-Performance Liquid Chromatography
ICF	Informed Consent Form
ICH	International Conference on Harmonization
ICP-MS	Inductively Coupled Plasma Mass Spectrometry
ICP-OES	Inductively Coupled Plasma Optical Emission Spectrometry
ICRP	International Commission on Radiological Protection

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IEC	Independent Ethics Committee
IL-6	Interleukine-6
IMP	Investigational Medicinal Product
INR	International Normalized Ratio
IRB	Institutional Review Board
ITLC	Instant Thin Layer Chromatography
MBq	Mega Becquerel
MIRD	Medical Internal Radiation Dose
MCV	Mean Corpuscular Volume
MedDRA	Medical Dictionary For Regulatory Activities
MRI	Magnetic Resonance Imaging
NSAID	Non-Steroidal Anti-Inflammatory Drug
NOAEL	No Observed Adverse Effect Level
PBS	Phosphate Buffered Saline
PI	Principal Investigator
PoC	Proof of Concept
PS	Phosphatidylserine
PT	Prothrombin Time
PTT	Partial Thromboplastin Time
PV	PharmacoVigilance
QA	Quality Assurance
QC	Quality Control
RA	Rheumatoid Arthritis
RBC	Red Blood Cell
RCP	Radiochemical Purity
ROI	Region of Interest
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SEC-HPLC	Size-Exclusion HPLC
SI	Sacro-Iliac
SOP	Standard Operating Procedure
SpA	Spondyloarthritis

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SPECT	Single-Photon Emission Computed Tomography
TBR	Target to Background Ratio
TLC	Thin Layer Chromatography
TNF	Tumor Necrosis Factor
US	United States
UV/Vis	Ultraviolet/Visible
WBC	White Blood Cell
WFI	Water For Injection
WHO	World Health Organization
VOI	Volumes Of Interest

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## 3 INTRODUCTION

### 3.1 Background and rationale

Spondyloarthropathy or spondyloarthritis (SpA) represents several types of autoimmune inflammatory arthritis which share a number of clinical and genetic features. These include; ankylosing spondylitis (AS), reactive arthritis, psoriatic arthritis or spondyloarthritis, SpA associated with inflammatory bowel disease (IBD), which includes Crohn's disease (CD) and ulcerative colitis (UC), undifferentiated SpA and juvenile-onset spondyloarthritis [Dougados 2011, Ehrenfeld 2012, Poddubnyy 2012].

There are certain characteristics linking AS and spondylitis associated CD together based on sharing some of the genetic, clinical, immunological, and microbial features [Ebringer A et al 2007]. Furthermore, most if not all SpA conditions are thought to have a fundamental link with the gut lesions and enterobacterial microbes [Rashid T et al 2011]. For example, around 10 percent of patients with AS have overt IBD, whilst 70% of AS patients have subclinical terminal ileitis [Mielants H et al 1995]. Axial and peripheral arthritis can occur in up to 30% of patients with CD [Orchard T et al 2009], and the prevalence of AS might increase to up to 6% in patients having CD [Palm Ø et al 2002]. Moreover, HLA-B27 positive patients with IBD were shown to have higher chance of developing AS compared to those without IBD [Wright W et al 1978].

Clinically patients present with sacroilitis and inflammatory back pain, peripheral arthropathy, enthesitis, extra-articular or extra-spinal involvement, including of the eye, heart, lung and skin in the absence of rheumatoid factor or subcutaneous nodules. SpAs also display a strong hereditary tendency and a varying association with HLA-B27 (and other less common genes) suggesting that these disorders share patho-physiologies [Wending 2013, Bleil 2014, Hreggvidsdottir 2014]. Overall SpAs have disease prevalence similar to rheumatoid arthritis (RA) in the general population [Dougados 2011, Ehrenfeld 2012, Poddubnyy 2012].

Challenges in the management of SpAs include; the prevention of new and/or progressive bone formation/spinal fusion and the lack of robust measures for early diagnosis [Poddubnyy 2012, Wending 2013, Hreggvidsdottir 2014]. The introduction of tumor necrosis factor (TNF)-alfa-blocking agents in the past decade has provided the first effective, though imperfect, disease-modifying anti-rheumatic drug therapy for SpA [Dougados 2011, Ehrenfeld 2012, Poddubnyy 2012]. However, as stated above these agents and potential new disease modifying drugs would be best used earlier in the disease course. In order to identify and treat patients with SpA early in the disease process, prior to radiographic changes of erosion and bony ankyloses of the sacro-iliac joints and spine, T1-weighted images alone or together with STIR images are considered of most importance for the MRI as indicative of SpA by the vast majority of radiologists and rheumatologists. For diagnosing SpA by MRI in the inflammatory back pain context, the T1-weighted sequence, either alone or in combination with STIR, still is considered as more relevant approach (Weber 2010).

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In this clinical trial, we propose to test the feasibility of  $^{99m}\text{Tc}$ -annexin V-128, an in vivo marker of phosphatidylserine (PS) expression [Blankenberg 1998 & 2009], to detect and assess the severity of inflammation at sites of disease in SpA patients. PS is an anionic membrane phospholipid present in all mammalian cells which is actively restricted to the inner leaflet of the lipid bilayer by ATP dependent pumps within the plasma membrane. However in conditions of severe metabolic stress, activation of the apoptotic cell death pathway or persistent or chronic neuronal excitation/depolarization PS is selectively exposed on the cell surface [Smith 2009, Kenis 2010]. Annexin V, an endogenous human intracellular protein specifically binds to membrane bound PS with a high affinity ( $k_d = 1$  to  $2$  nM) permitting its use as an imaging agent of cellular stress, depolarization and apoptosis both in vitro and in vivo [Tait 2005, 2006].

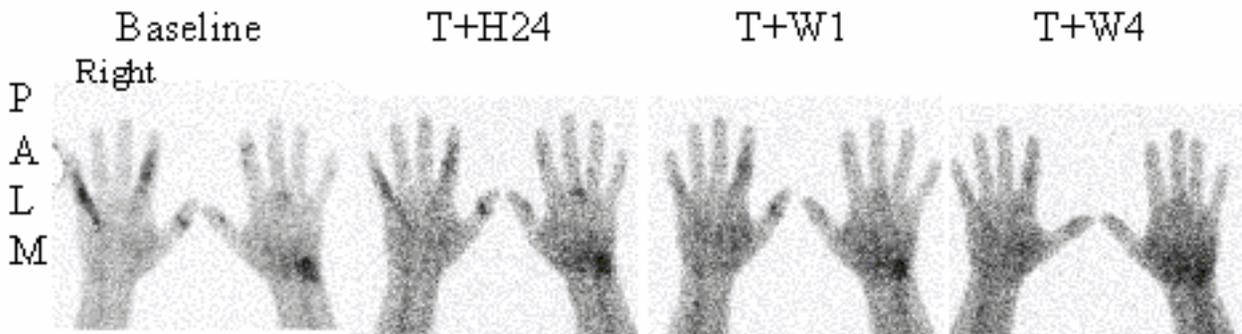
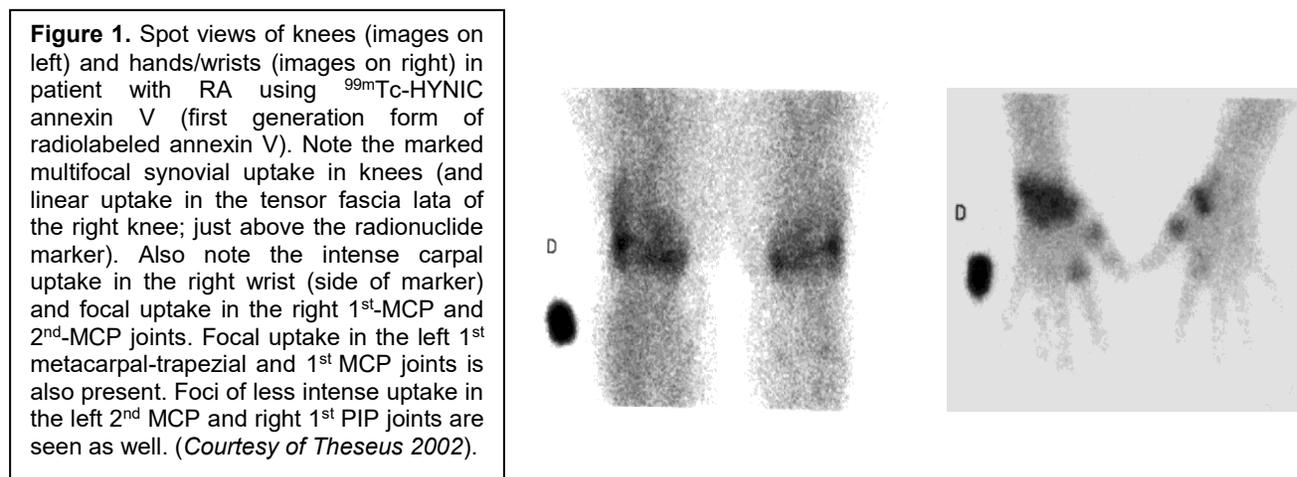
**Preliminary data with  $^{99m}\text{Tc}$ -annexin V for imaging of acute and chronic inflammation.**

Radiolabeled forms of annexin V, including  $^{99m}\text{Tc}$ -HYNIC-annexin V and  $^{99m}\text{Tc}$ -annexin V-128 have shown the ability to image and monitor sites of autoimmune and infectious inflammation and response to treatment in multiple animal models and humans [Vriens1998, Blankenberg 2000, Ogura 2000 & 2001, Tokita 2003, Lorberboym 2009, Benali 2014, Hardy 2014]. The ability of annexin V to localize sites of inflammation is directly related to its highly specific binding to PS, whose exposure on the cell surface is an indicator of cellular stress that if unchecked can lead to apoptosis (programmed cell death) and recognition by the immune system for removal. In general PS externalization due to ongoing stress (as opposed to apoptosis) demonstrates far lower levels of PS exposure as compared with apoptosis and other forms of cell death (Hamill 1999, Geske 2001). These relatively low levels of PS exposure can be readily counteracted by the prompt removal of the offending physiologic stressor and is referred to as “reversible PS expression”. However, if the stress(ors) remains uncorrected, a cell may undergo apoptosis. Reversible uptake of radiolabeled annexin V has been observed in vitro and in human models of forearm muscle exercised induced ischemia (Riskin 2005 & 2006, Rongen 2005). The ability of annexin V to bind to cells with low but potentially reversible levels of PS exposure may also explain the uptake of radiolabeled annexin V outside (contralateral) regions of apoptosis in patients with hypoxic-ischemic reperfusion injury in the heart (Thimister 2003, Sarda-Mantel 2006) or brain (Lorberboym 2006, Blankenberg 2006, Tang 2007). In fact, even in regions with apoptosis/ischemic injury, there are far more annexin V–positive cells after the administration of radiolabeled annexin V than apoptotic nuclei as seen by TUNEL staining (Tang 2007). These observations suggest that much (if not most) uptake of annexin V after ischemic reperfusion injury may be due to large numbers of stressed cells (not necessarily committed to apoptosis) with relatively low levels of PS expression in contrast to the relatively fewer cells with high levels of PS exposure that are irreversibly committed to apoptosis. The ability of annexin V to localize cells that are stressed but not necessarily committed to apoptotic cell death suggests that this radiotracer can be used to identify tissues or organs at risk for irreversible injury, such as seen in hypoxic-ischemic injury (Taki 2007) or chronic heart failure (Kietselaer 2007), or sites of active disease that are seen in infection (Rouzet 2008, Kietselaer 2009), unstable atherosclerotic plaques (Tahara 2009), allograft rejection (Narula 2001), or autoimmune disorders such as SpA (Tokita 2003, Perker 2004). Annexin V imaging could therefore be useful in the serial assessment of acute and chronic disorders in organs or tissues at risk for permanent damage in which prompt treatment with effective drug or surgical intervention

may prevent irreversible cellular injury and cell death.

### Brief history of <sup>99m</sup>Tc-HYNIC-annexin V in patients with rheumatoid arthritis or infection

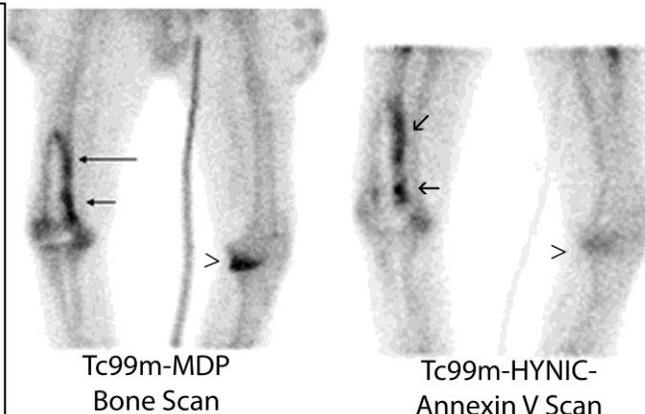
<sup>99m</sup>Tc-HYNIC-annexin V, the first generation form of radiolabeled annexin V to be tested in humans starting in 2000, was effective at imaging disease within the knees and wrists/hands of several rheumatoid arthritis (RA) patients and was also able to monitor the early effects of biologic modifying therapy at one week in a small pilot study performed in Liege, Belgium in 2002 (Figures 1 & 2). The sensitivity of <sup>99m</sup>Tc-HYNIC-annexin V for regions of low grade chronic inflammation due to subacute bacterial infection of hip and knee prostheses was confirmed in a pilot study in Israel (Figure 3). Interestingly reactive bone formation, osteoarthritis and prosthetic joint loosening while positive on conventional bone scans with <sup>99m</sup>Tc-MDP did not result in significant increases in annexin V uptake. While effective in human trials <sup>99m</sup>Tc-HYNIC-annexin V due to the metabolism of the HYNIC linker had relatively high nonspecific tracer uptake in bone marrow and renal tubular cells prompting the development of the second generation form of self-chelating radiolabeled annexin V known as <sup>99m</sup>Tc-annexin V-128.



**Figure 2.** Spot views of the hands/wrist of another patient with RA with injection and radionuclide imaging (1 hr later) with <sup>99m</sup>Tc-HYNIC annexin V performed at baseline and at 24 hrs, one week and four weeks after starting biologic modifying therapy with chimeric anti-TNF monoclonal antibody (Infliximab™). At baseline intense linear focal uptake was noted in the right 2<sup>nd</sup> and 5<sup>th</sup> flexor tendon sheaths and left triquetral-pisiform joint space. Less intense focal uptake was noted in the 1<sup>st</sup> PIPs bilaterally and left 2<sup>nd</sup> DIP, and 3<sup>rd</sup> MCP joints. There was a prompt significant decrease in focal uptake 24 hours after the start of Infliximab treatment (i.e. uptake decreasing to soft tissue background) particularly of the tenosynovitis of the right hand. At one week there were no signs of tenosynovitis remaining though there was persistent low diffuse uptake within the left triquetral-pisiform joint. At four weeks there were no sites of focal increased uptake in either hand or wrist though there was a persistent low diffuse uptake of tracer in the region of the left carpus just above soft tissue background. (Courtesy of Theseus Corp. 2002).

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**Figure 3.** Bone scan of the knees (image on left) and corresponding  $^{99m}\text{Tc}$ -HYNIC annexin V scan (image on the right) several days later in a 76 year old man with a painful right knee prosthesis and signs of loosening on plain x ray. Arrows on the medial side of the femoral portion of the knee prosthesis mark intense focal linear uptake both on bone scan and on the annexin scan. This region of uptake corresponded to the infected portion of the prosthesis at surgery. Also note that annexin V uptake was highly specific as compared to bone scan in which focal non-specific uptake of Tc99m-MDP was present over the entire femoral portion of the prosthesis as well as the osteophytic spurring/osteoarthritis of the medial aspect of the right knee (proximal tibia) as indicated by arrowheads. *From: Lorberboym M, et al. J Nucl Med 2009; 50:534–537.*



### 3.2 $^{99m}\text{Tc}$ -rhAnnexin V-128 Risk-Benefit Assessment

Preclinical characterization of  $^{99m}\text{Tc}$ -rhAnnexin V-128 safety profile indicated that the product can be safely administered, both from the point of view of the proposed radioactive dose ( $350 \pm 10\%$  MBq) and also considering the total drug substance (protein) present in a  $350 \pm 10\%$  MBq dose (0.4 mg). In particular, the radiation safety assessment performed based on extrapolations from animal biodistribution and dosimetry data showed that, for the proposed injected dose of  $350 \pm 10\%$  MBq, the estimated kidney absorbed dose in human is around 60 mGy, which is below the level recommended by FDA for the single dose per organ (0.05 Sv), as generally recognized as safe in the FDA Guidance for Industry and Researchers (2010) and considerably lower than the threshold for nephrotoxicity reported in literature, that is 14 Gy (corresponding to 14 Sv for  $^{99m}\text{Tc}$ ) when delivering radiation dose in 2 fractions, as reported by Emami et al., 1991, for tolerance dose 5/5 (the probability of 5% complications within 5 years of irradiation) reported in literature [30]. These estimated kidney absorbed doses are also lower than those achieved with some technetium base diagnostics currently used in the nuclear medicine practice (e.g.  $^{99m}\text{Tc}$  MoAb antiCEA has kidney as critical organ with an absorbed dose of 100-400 mGy for an injected dose of 1000 MBq).

The estimation of effective dose for total human body, from an injected dose of  $350 \pm 10\%$  MBq, is  $2.2 \pm 10\%$  mSv, that is far below the FDA single-study limit of 30 mSv for research subjects, as per FDA Guidance for Industry and Researchers, 2010.

Therefore, animal data from biodistribution/dosimetry studies and extrapolation of human kidney absorbed and total body effective dose, along with the current nuclear medicine practice concerning technetiated diagnostics, support the proposed single dose of  $350 \pm 10\%$  MBq of  $^{99m}\text{Tc}$  Annexin V-128 for the Phase II study.

Regarding the toxicity profile of the non-radiolabeled protein, a package of pre-clinical studies was

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designed in order to investigate the potential toxicity of rhAnnexin V-128. Preliminary 7-day toxicity studies conducted in rats and dogs, showed that rhAnnexin V-128, administered intravenously once daily, was well tolerated up to the highest tested dose, that is 450 fold (in rats) and 200 fold (in dogs) the proposed dose in humans.

The main repeated-dose toxicity studies were conducted in rats and dogs as well, and consisted of 2-week daily intravenous administrations of rhAnnexin V-128, at doses that were approximately 25, 50 and 100 times the intended dose in humans. The toxicity profile was in general favorable, with some treatment-related findings in the spleen (germinal centers hyperplasia) and in the liver (periportal eosinophilic cell infiltration) of the rats at the intermediate and high dose, and some periportal subchronic inflammation in dog liver at the high dose. These signs were in general reversible after 2 weeks recovery period. The No-Observed-Adverse-Effect-Level (NOAEL) was defined to be 0.2 mg/kg/day in rats (25 fold the intended dose in humans) and 0.4 mg/kg/day in dogs (50 fold the intended dose in humans).

These findings meet the safety margins recommended in the relevant guideline, which indicates that the dose intended to be used in Phase I studies should be at least 100 times lower than the NOAEL found in the acute toxicity studies in the rat and at least 25 times lower than the NOAEL found in the repeated toxicity studies in rat and dog.

As described in the Investigator's Brochure, it is worth noticing that no adverse reaction, including any sign of allergic or anaphylactic reaction, was reported in animals during in vivo phase of any of the toxicity studies performed in rats and dogs. As a reference the dose range tested in animals was within 25 to 450 times the intended human dose).

In addition to the in vivo toxicology studies mentioned above, the safety of rhAnnexin V-128 was evaluated in a whole blood cytokine release ex-vivo assay. The results showed that rhAnnexin V-128 did not induce any cytokine release.

An open label, single dose, Phase I clinical study of safety, tolerability, pharmacokinetics and nuclear medicine imaging of <sup>99m</sup>Tc-rhAnnexin V-128 intravenous administration of 350 MBq single intravenous bolus in 12 healthy adult volunteers has been conducted. A series of images for up to 24 hours from the time of injection and blood samples were obtained from time of screening, dosing day and 24 hours after injection, with follow up visits at 72 hrs, 7 days and 30 days post injection. A physical examination was conducted 72 hrs, 7 days and 30 days post administration of <sup>99m</sup>Tc - rhAnnexin V-128. No abnormal findings have been noted after administration of the studied imaging product and during the observation period. No immunology response has been reported in the immunogenicity assessment 14 and 30 days post administration.

The concentration-time profiles for total radioactivity in blood and serum were similar at the injected dose. Concentrations of radioactivity were measurable in blood and serum of all subjects between 5 min and 24 h, reaching the higher concentration of about 30 ngEq/mL at the first sampling time. Blood and serum area under the concentration-time curves (AUC) were highly correlated (Pearson  $r = 0.77$ ;  $p=0.006$ ), and the blood-to-serum AUC ratio averaged 0.72 (range, 0.65 to 0.82)

suggesting moderate distribution of rhAnnexin V-128-related material into blood cells. Mean terminal  $t_{1/2}$  of radioactivity averaged 757 min in blood and 766 min in plasma (Pearson  $r = 0.92$ ;  $p=0.0001$ ), further suggesting that serum is a suitable matrix for the representation of rhAnnexin V-128 pharmacokinetics in whole blood.

The mean serum concentrations of rhAnnexin V-128 based on ELISA analysis were lower and disappeared faster than the mean serum concentrations of total radioactivity. However, the ELISA assay was generally not sensitive enough to follow serum concentrations for more than 90-180 min, this rapid decay probably approximating an initial phase of the compound distribution to tissues. Thus, an elimination phase could not be clearly defined for rhAnnexin V-128 at this intravenous dose. Estimates of pharmacokinetic parameters for rhAnnexin V-128 suggest low to moderate mean total clearance (Cl; 45.4 mL/min) and moderate to large mean steady-state volume of distribution (Vss; 49.8 L) in healthy volunteers. Inter-subject variability of pharmacokinetic parameters was moderate to high with coefficients of variation (C.V.) ranging from about 38% for systemic Cl to about 70% for Vss.

Around 65% (range, 36 to 81%) of the administered radioactive dose was eliminated within 6 h post-dosing, and only about 16% (range, 3% to 22%) and 3% (range, 1.4% to 4.5%) were recovered during the subsequent 6-18 h and 18-24 h collection periods, respectively, in eleven subjects (one subject had incomplete urine collection). The overall dose of administered  $^{99m}\text{Tc}$ -rhAnnexin V-128 recovered in urine during the 0-24 h post-dose interval was 85% (61% to 114%), in the eleven evaluable subjects. There was negligible excretion of radioactivity in feces during the 0-24 h period, which accounted for a mean of only about 1% (range, 0.1% to 3.5%) of the administered dose.

## 4 STUDY OBJECTIVES AND HYPOTHESES

This Phase II study will be undertaken to determine the presence and degree of  $^{99m}\text{Tc}$ -rhAnnexin V-128 uptake at sites of clinical disease in patients with clinical suspicion or confirmed diagnosis of SpA. While the majority of these patients will be diagnosed with AS, there will be a substantial number of patients diagnosed with other types of disease including IBD related SpA and Psoriatic arthritis. We hypothesize that SpA patients with active disease as manifested clinically by dull low back, buttock or hip pain will have significant increased  $^{99m}\text{Tc}$ -rhAnnexin V-128 uptake in the sacroiliac joints and/or spine which will correlate with both clinical scoring and changes with T1 and STIR without contrast MRI findings. We also hypothesize that we will be able to directly quantify the severity of inflammation at sites of disease with a higher degree of specificity and sensitivity than MRI, CT and plain film radiography.

Eligible patients after obtaining informed consent, baseline blood labs, and urinalysis, will be injected once with  $^{99m}\text{Tc}$ -rhAnnexin V-128 followed by SPECT/CT imaging of the lumbosacral spine, sacroiliac joints and abdomen at 60 min and 2 hrs after injection of the radiotracer. Patients will undergo also a whole body planar scan and spot views on specific areas at the same time-points (spot views may not be repeated at T+2hrs). An optional whole body planar scan will be done 24hrs post injection for 5 patients as well as spot views, if deemed appropriate by the investigator.

**Study Protocol**

Knowledge of the expected dynamic range of the disease related changes in  $^{99m}\text{Tc}$ -rhAnnexin V-128 uptake in correlation with clinical measured of disease severity within affected joints will be critical for the power calculations and sample size estimates as part of the design of a follow up studies in SpA patients in order to further demonstrate the benefit of this new imaging procedure versus conventional imaging with MRI and plain film radiography. Furthermore, it is expected that the quantitative capabilities of SPECT/CT and the inherent sensitivity of  $^{99m}\text{Tc}$ -rhAnnexin V-128 imaging and reduced cost will replace MRI as a screening tool for the work up of patients with SpA.

As secondary goal, we also wish to determine the ability (specificity) of  $^{99m}\text{Tc}$ -rhAnnexin V-128 to distinguish between active autoimmune arthritis versus osteoarthritis particular of the lumbosacral spine and SI joints of patients with SpA. We also hypothesize that some of SpA patients with active disease have subclinical Crohn's disease. Therefore, we wish to perform a pilot study on the localization of  $^{99m}\text{Tc}$ -rhAnnexin V-128 in the abdomen.

### **4.1 Study objectives**

Primary Objective:

- To determine the magnitude and dynamic range of  $^{99m}\text{Tc}$ -rhAnnexin V-128 uptake in disease affected sacro-iliac or lumbar spine joints in patients with clinical suspicion or confirmed diagnosis of SpA.

Secondary Objectives:

- To determine clinical utility of  $^{99m}\text{Tc}$ -rhAnnexin V-128 SPECT imaging in the identification of chronic osteoarthritic active sites of SpA patients compared with MRI in patients with disease-affected sacro-iliac or lumbar spine joints.
- To assess the presence of antibodies against rhAnnexin V-128 at baseline and post-treatment
- To determine the localization pattern and magnitude of focal uptake of  $^{99m}\text{Tc}$ -rhAnnexin V-128 within the abdomen in patients with clinical suspicion or confirmed diagnosis of Spondyloarthritis (SpA).

### **4.2 Study hypotheses**

We hypothesize that  $^{99m}\text{Tc}$ -rhAnnexin V-128; a second generation form of  $^{99m}\text{Tc}$  radiolabeled annexin V with significantly higher PS binding affinity and improved in vivo localization properties, will be highly specific in imaging degenerative osteoarthritis particularly of the lumbosacral spine

**Study Protocol**

and sacroiliac joints as seen by SPECT/CT in patients with SpA. We also hypothesize that  $^{99m}\text{Tc}$ -rhAnnexin V-128 will be localized within the abdomen of SpA patients with active disease showing suspicious Crohn's disease.

### ***4.3 Study justification***

At the completion of this study, we will have preliminary data on the expected magnitude and dynamic range and specificity of  $^{99m}\text{Tc}$ -rhAnnexin V-128 uptake at sites of active disease in SpA patients at presentation.

## **5 STUDY DESIGN**

### ***5.1 Study Outline***

This is a monocentric, open label, PoC, Phase II study. Patients who have signed the informed consent and are eligible for study participation according to the inclusion and exclusion criteria will receive a single intravenous bolus of  $^{99m}\text{Tc}$ -rhAnnexin V-128 on Day 0 followed by SPECT/CT of the sacro-iliac joints, spine and abdomen at 60 min and 2hrs after injection of the radiotracer within 14 days after screening in order to minimize the potential delay in changing NSAID therapy or non-biologic DMARD or the start of non-biologic DMARD or biologic DMARD (excepted for the first 5 enrolled patients who will not be eligible for the start of biologic DMARD). Patients will undergo also a whole body planar scan and spot views on specific areas at the same time-points (spot views may not be repeated at T+2hrs). We will also perform T1 and STIR without contrast MRI of the sacro-iliac joints and lumbar spine within this same time window,

Details on study assessments are provided in Protocol Section 8 and Table1.

### ***5.2 Study duration and End of Study***

Study duration is maximum 7 weeks (including the screening period) for each patient. The end of the study is defined as the moment that the last enrolled patient has completed the planned assessments at Day 30.

## **6 PATIENT SELECTION**

Approval for the study will be obtained from the local IRB of Cedars-Sinai Medical Center Office of Research of Compliance. All patients will provide written informed consent prior to the initiation of any study procedures.

### ***6.1 Inclusion Criteria***

**For the first 5 patients enrolled in the POC part:**

1. Patients with clinical suspicion or confirmed diagnosis of SpA, based on the ASA criteria with active symptoms including back, hip or buttock pain prior to:

**Study Protocol**

- A change in NSAID therapy or
- A change in non-biologic DMARD or
- A start of non-biologic DMARD.

**For the next 15 patients enrolled in the Phase II part:**

1. Patients with clinical suspicion or confirmed diagnosis of SpA, based on the ASAS criteria with active symptoms including back, hip or buttock pain prior to:
  - A change in NSAIDs therapy
  - A change in non-biologic DMARD
  - A start of non-biologic DMARD
  - A start of biologic DMARD

**For all patients:**

2. Age over 18 years old.
3. Signed Informed Consent Form

**6.2 Exclusion Criteria**

1. Pregnancy or lactation
2. Liver impairment (ALT, AST or Bilirubin > 2 ULN) at screening visit or baseline
3. Kidney impairment (serum creatinine > 1.5 mg/dL)
4. History of any disease or relevant physical or psychiatric condition or abnormal physical finding which may interfere with the study objectives at the investigator judgment
5. Known hypersensitivity to the investigational drug or any of its components
6. Contraindication(s) to the MRI procedure (claustrophobia, prosthetic valve, pacemaker, inability to lie still in a supine position)
7. Participation to another clinical trial within 4 weeks before study inclusion except for patients who have participated or who are currently participating in an interventional study without any study drug administration.

**6.3 Number of patients**

Up to twenty evaluable patients will be recruited.

**6.4 Patients Identification**

Each patient will be anonymized and identified with a patient ID number. A unique patient identification number (Patient ID) will be assigned at the start of the screening period to each patient who signs the informed consent form until the study termination of the patient. This number will identify the subject throughout the study. Patient IDs will include the 2-digit protocol number (03), the 2-letter country code (US) and a 3-digit patient number (ex: 03-US001 for first subject in).

**Study Protocol****6.5 Premature Discontinuation**

The withdrawal of a study patient is mandatory in the following cases:

- Pregnancy
- Protocol violation determined as critical
- Lost to follow-up
- Serious intercurrent illness or other safety reasons for what the Investigator considers it is in the best interest of the participant to withdraw from the study
- Screening failure

A “screening failure” is a patient who has signed the informed consent, but who does not meet all selection criteria following the screening evaluations. For patients not considered eligible after the screening period, the reason for not being eligible must be documented. No additional assessments are needed. Patient information collected at the Screening visit will be entered in the e-CRF and will be used in the study analysis.

Patients who will not have received the study product will be replaced.

The patient is free to withdraw from the study for any reason and at any time without giving reason for doing so and without penalty or prejudice. It is also possible that the Sponsor or the regulatory authorities request termination of the study if there are concerns about conduct or safety.

The primary reason for a patient’s withdrawal from the study should be determined as possible. The date and reason for discontinuation must be documented in the e-CRF.

**6.6 Prohibition and Restrictions**

The most important required restriction for study patients is pregnancy. Pregnancy tests will be performed for all patients (of child bearing potential) at baseline and prior to the radiotracer administration.

Imaging is performed ambulatory. After imaging, patient may go back home or go back to work without particular precaution for him or his relatives.

Any concomitant medication deemed necessary for the welfare of the subject during the study may be given at the discretion of the Investigator. However, it is the responsibility of the Investigator to ensure that details regarding the medication are recorded in full in the case report form, including any changes that have occurred during the study.

**7 STUDY MEDICATION AND TREATMENT****7.1 Packaging and Labelling**

The full name of the Investigational New Drug is “Kit for the Preparation of Tc-99m Recombinant Human Annexin V-128 for Injection”. The kit will be prepared, packaged and released according to Sponsor Standard Operating Procedures (SOPs), and in compliance with the principles of the Good Manufacturing Practice (GMP) guidelines, ICH Good Clinical Practice (GCP) guidelines, and

applicable federal, state and local laws/regulations.

The investigational product will be supplied by the Sponsor as a sterile, single vial lyophilized kit for reconstitution with  $^{99m}\text{Tc}$  solution.

A single dose vial contains an amount of rhAnnexin V-128 and excipients suitable for preparing up to 740 MBq of injectable  $^{99m}\text{Tc}$ -rhAnnexin V-128. The excipients in the kit are the following: stannous chloride (reducing agent), sodium  $\alpha$ -D-Glucoheptonate dihydrate (transchelating agent), gentisic acid sodium salt hydrate (radiation stability enhancer), hydroxypropyl- $\beta$ -cyclodextrin (solubilizer), sodium metabisulfite (antioxidant) and trehalose dihydrate (lyoprotectant and cake-forming agent). Lactic acid is also present as buffering agent.

The reconstitution procedure and the specifications of the kit and of the radiolabeled imaging agent are described in Appendix I.

## **7.2 Handling of Study Medication**

$^{99m}\text{Tc}$ -rhAnnexin V-128 must be administered at the investigational site. The study medication must be stored, handled and administered only by qualified/authorized personnel and must be prepared in accordance with pharmaceutical quality requirements, and radiation safety regulations.

Based on the stability tests performed (Appendix I), the Annexin Kit can be stored upon receipt at  $5^{\circ}\text{C} \pm 3^{\circ}\text{C}$  until the expiry date stated on the labels.

A single dose vial containing 0.4 mg of lyophilized rhAnnexin V-128 will be reconstituted with 2 mL  $\pm$  0.2 mL containing 740 MBq  $\pm$  74 MBq (20 mCi  $\pm$  2 mCi) of  $^{99m}\text{Tc}$  from the generator. This amount of radioactivity provides a sufficient amount of radioactivity for QC testing and the foreseen  $^{99m}\text{Tc}$ -rhAnnexin V-128 dose of  $350 \pm 10$  % MBq for up to 4 h after labelling. The administration volume (corresponding to  $350 \pm 10$  % MBq) is calculated according to the estimated time of injection, on the basis of the physical decay of the radionuclide (half-life = 6.02 h). Stability studies have demonstrated radiochemical purity at 6 h from labelling greater than 90%, which is in line with current specifications of the Kit for the preparation of  $^{99m}\text{Tc}$ -rhAnnexin V-128 for injection. For the purpose of this study, it is recommended to administer the reconstituted solution within 4 hours after completion of the labelling reaction. It is also required to determine the amount of radioactivity injected to the patient by measuring the radioactivity before and after injection with an appropriate radioactivity calibration system.

## **7.3 IMP Packaging and Storage**

The site personnel will maintain shipping, dispensing, and collection logs. All investigational product will be stored and inventoried according to the protocol instructions and applicable federal, state, and local regulations and will be stored in a secure, locked location with limited authorized access at the investigational site.

## **7.4 Precautions and recommendation for use**

Technetium-99m eluate should be obtained from a commercially available Mo-99/Tc-99m generator, approved by regulatory authorities, that has been eluted within the past 24 hrs. The radiopharmacist will be directed to use the eluate within one hour of milking the generator.

On ongoing basis the Investigator/radiopharmacist agrees to conduct a study medication supply inventory and to record the results of this inventory on the IMP Reconciliation Form. It must be possible to reconcile delivery records with those of used and unused medication. Any discrepancies must be accounted for. Appropriate forms of deliveries and returns must be signed by the responsible person.

Unused "Kits for preparation of  $^{99m}\text{Tc}$ -rhAnnexin V-128 for injection" will be locally discarded after monitoring by the Clinical Research Associate (CR A).

Refer to Appendix I for detailed instruction on the Radiolabeled Imaging Product  $^{99m}\text{Tc}$ -rhAnnexin V-128, Cautionary notes, Analytical and Biological Controls, Stability and Shelf Life.

### **7.5 Study Medication Dose, Mode of Administration, Batch Number**

The "Kit for preparation of  $^{99m}\text{Tc}$ -rhAnnexin V-128 for injection" consists of 1 patient dose. The single dose vial (0.40 mg rh-Annexin V-128) will be reconstituted with 2 mL  $\pm$  0.2 mL containing 740 MBq  $\pm$  74 MBq (20 mCi  $\pm$  2 mCi) of  $^{99m}\text{Tc}$ .

The labelling reaction requires 90 minutes and the reconstituted radiolabeled product is stable for 6 h. For the purpose of this study, it is recommended to administer the reconstituted solution within 4 hours after completion of the labelling reaction. Patients will receive a single administration of  $^{99m}\text{Tc}$ -rhAnnexin V-128 on Day 0. When the investigational product is received at the site, the investigator, pharmacist or authorized designee shall check for accurate delivery and acknowledge receipt by signing and dating the documentation provided by the Sponsor and returning it to the Sponsor or designee. A copy of this documentation shall be retained for the Investigator file.

The dispensing of the investigational product shall be carefully recorded on an investigational product accountability form provided and an accurate accounting must be available for verification by the CRA at each monitoring visit.

Unused investigational product must not be discarded or used for any purpose other than the present study. The CRA will periodically collect the Drug Accountability Forms and will check all investigational product (both unused and used) before making arrangements for authorizing their destruction at the investigational site.

## **8 ASSESSMENTS**

Voluntary written informed consent will be obtained prior to the initiation of any study-related procedures. Study conduct procedures will include screening, the imaging visit and follow-up visits.

### **8.1 Baseline Assessments**

- Each patient's demography: gender, ethnicity, medical history, and relevant baseline characteristics will be recorded at Screening visit.

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- Inclusion/exclusion criteria will be checked at Screening.
- Women of childbearing potential must have negative urine pregnancy test at Screening before the radiotracer administration and before MRI (in case the MRI is not performed on the same day of the screening visit).
- Vital signs will be taken at Screening.
- Physical examination will be conducted at Screening.
- BASDAI, BASFI, BASMI and MASES assessments will be performed.

### 8.2 Safety Assessments

- Physical examination will be conducted at D30
- Measurement of vital signs (systolic and diastolic blood pressure and heart rate) will be performed 15 minutes before the radiotracer administration and at the end of the last imaging procedure at Day 0.
- Women of childbearing potential must have negative urine pregnancy test before injection of the radiotracer at Day 0.
- All medications taken from 2 weeks prior to the administration date through the end of study are to be recorded as prior and concomitant medications, including therapies for SpA.
- Any adverse events occurring during the course of the study will be reported and recorded in the eCRF. 24 hours after treatment injection (at Day 1), each patient will be called in order to inquire if they experienced signs or symptoms such as edema, rash or have any clinical findings.

### 8.3 Standard Laboratory Assessments

General laboratory analysis will be performed and blood samples will be collected at screening and at Day 30.

Blood sample will be drawn and urine will be collected as mentioned in the study chart. A total volume of 50 mL, including blood sample for immunogenicity assessment, will be collected during the study for each patient.

Hematology	Coagulation	Blood Chemistry	Urinalysis
<ul style="list-style-type: none"> <li>• WBC with differential</li> <li>• RBC</li> <li>• Platelets</li> <li>• Hb</li> <li>• MCV</li> <li>• Hematocrit</li> </ul>	<ul style="list-style-type: none"> <li>• PT</li> <li>• PTT</li> <li>• INR</li> </ul>	<ul style="list-style-type: none"> <li>• BUN</li> <li>• Serum creatinine</li> <li>• Uric acid</li> <li>• Albumin</li> <li>• Total bilirubin</li> <li>• Direct bilirubin</li> <li>• AP</li> <li>• AST/ASAT</li> </ul>	<ul style="list-style-type: none"> <li>• Dipstick test<sup>1</sup></li> <li>• Pregnancy test <i>(at screening and before injection of study product)</i></li> </ul>

		<ul style="list-style-type: none"> <li>• ALT/ALAT</li> <li>• Gamma-GT</li> <li>• Sodium</li> <li>• Potassium</li> <li>• LDH</li> <li>• CRP</li> <li>• Calcium</li> <li>• Glucose</li> <li>• Chloride</li> <li>• Phosphorus</li> <li>• Total cholesterol</li> <li>• Total protein</li> </ul>	
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<sup>1</sup>In case one of the assessments of the dipstick test is positive, a microscopy analysis of the urine must be performed.

### **8.4 Clinical assessment**

A clinical evaluation including the search for enthesopathy – using the Maastricht Score (MASES) - and assessments with BASDAI, BASFI and BASMI will be performed at Screening.

The clinical assessment will be completed by the measurement of biological markers of inflammation (CRP and CBC) at Screening and at Day 30.

### **8.5 Imaging**

Patients will be injected intravenously with 350 ± 10 % MBq of <sup>99m</sup>Tc-rhAnnexin V-128 with an 18 gauge angiocath IV and saline TKO drip set up placed in the antecubital fossa.

Bone Lumbar Spine SPECT/CT, SI SPECT/CT and abdomen SPECT/CT will be performed 60 minutes and 2 hours after injection of radiotracer followed by a whole body planar scan and spot views on specific areas at the same timepoints (spot views may not be repeated at T+2 hrs if deemed unnecessary)

The Infinia™ Hawkeye 4 SPECT/CT (GE Healthcare) scanner was used for patients with the following parameters; start at 0°, 180° rotation/detector, 64 steps, 3° per step, 30 sec per step, overall ARC 360°, (scan time = 23 minutes) 128 x 128, multi-purpose collimator (Skylight) in 1 x zoom mode. An attenuation and diagnostic CT scan will be performed on the lumbar spine using a 512 x 512 matrix, 140 keV, 2.5 mA, axial 1 sec, 5 mm slices.

Semi-quantitative assessment of inflammatory joints and abdomen will be performed in terms of TBR as well as by calculating the relative uptake in ROI/VOIs. ROI and VOI defined and use in the different scintigraphies will strictly have the same pixel size and localization; and will be measured in counts.

In case a suspected anomaly is found at imaging, it would be reported to the rheumatologist and the patient's general practitioner.

A whole lumbar spine and sacro-iliac joint MRI will be performed within 14 days of <sup>99m</sup>Tc-annexin V-128 scanning using the following parameters: **SI Joint Sequences** /3 plane localizer / Whole pelvis

Coronal T1/ Whole pelvis Coronal T2 FS. Small FOV over SI Joints (~20 cm FOV): Sequences /Obl Axial T1. Obl Axial T2 FS (May use STIR or IDEAL for poor FS), Obl Coronal T1, Obl Coronal T2, FS (May use STIR or IDEAL for poor FS). Following pre/post sequences will be used to help assess for subtle sacroilitis: Obl Ax T1 FSE FS Pre / Obl Ax T1 FSE FS Post /Obl Cor T1 FSE FS Post. FOV notes: Small FOV width should be approximately from femoral head to femoral head; Oblique axial and coronal are prescribed relative to the sacrum. **Protocol for Spine:** Sag T1, T2 FSE no fat sat, STIR, FOV 28 / Ax T1 and T2 FSE (no fat sat), FOV 20.

## **8.6 Immunogenicity assessments**

As rhAnnexin-V 128 is an exogenous protein, development of antibodies against the protein must be assessed in order to evaluate the risk and consequences of such development.

Assays for anti-rhAnnexin V-128 IgG and IgM antibodies will be performed in serum samples by ELISA at screening visit. An additional sample is required at Day 30.

For this purpose 10 mL blood samples will be collected at screening and Day 30. At each time-point, after centrifugation, serum will be divided into six aliquots and frozen (-80°C). Three out of six serum aliquots will be then shipped to the central laboratory at [REDACTED]. Remaining samples will be shipped only if required for analysis and destroyed at study end if not required. Blood Samples Handling will be detailed in the Laboratory Manual provided by [REDACTED].

## **9 DATA MONITORING COMMITTEE (DMC)**

The DMC consists of representatives of investigators and Sponsor representatives and could include external person such as independent experts, if judged appropriate by the Sponsor. The main function of the committee will be to review the images of the first 5 patients and determine if there is a premature high evidence of the presence of inadequate technical performance in this patients group. The DMC will also determine if there is a high evidence of an excess of adverse events.

The DMC will review and evaluate the images of the first 5 patients in terms of image quality visual efficacy (uptake) and clinical relevance of the study product. The DMC will also review the examinations of safety data. The DMC will promptly give recommendations to continue or terminate the study. The DMC report will be edited by the Sponsor and sent to the ethics committee as well as competent authorities.

Preliminary results can be used by the Sponsor for publication purpose, before the end of the main trial.

## **10 STATISTICAL CONSIDERATIONS**

The statistical analysis of the present study will be performed in accordance with the principles stated in the Consensus-Guideline E9 (Statistical Principles for Clinical Trials) of the International

Conference on Harmonization (ICH).

Statistical method will be described in details in the statistical analysis plan (SAP). The considered approach is summarized here after.

### **10.1 General Statistical considerations**

If not stated otherwise, all statistical analyses will be conducted as follows.

Categorical data will be presented as frequencies, percentages and 95% confidence interval if relevant. For continuous data, N, missing, mean, standard deviation, median, quartiles, minimum, maximum and if relevant 95% confidence interval will be presented. Graphs such as box plots, circular diagrams or histograms can also be computed if relevant. p-values of 0.05 or lower will be considered as statistically significant. No adjustment will be made for multiple testing and missing data will not be replaced. Considering the small sample size planned in this study, non-parametric approach will be favored.

### **10.2 Sample size**

The number of patients in this study is not based on statistical power considerations. The planned sample includes 20 patients which is the number believed to provide sufficient data to assess the potential of <sup>99m</sup>Tc-rhAnnexin V-128 to image disease severity in SpA patients and provide sufficient preliminary data to plan larger pivotal trials focusing on the confirmation of the benefit for this new imaging procedure for the early diagnosis of SpA.

At first, five patients will be recruited and analyzed as a Proof of Concept (PoC). In the PoC part of the study, medical images will be reviewed by the DMC in charge of assessing the imaging potential of <sup>99m</sup>Tc-rhAnnexin V-128 in terms of image quality, uptake and medical relevance. After reaching a consensus, the committee will decide if the clinical investigation should continue with the remaining 15 patients. The planned number of patients in the PoC study is not based on statistical considerations. However, this number of patients should provide sufficient relevant information for the evaluation of the images in terms of quality and medical relevance.

### **10.3 Demographics and Other Subject Characteristics**

Demographic and other baseline data will be summarized descriptively. All background and demographic/SpA diagnostic data will be listed in detail.

### **10.4 Concomitant Medications**

Concomitant medications (including treatments for the underline disease) will be coded using World Health Organization (WHO) dictionary. Type and incidence of concomitant medications will be tabulated (generic terms).

### **10.5 Laboratory Tests**

Descriptive statistics including shift tables will be generated for all laboratory tests performed i.e. the

actual values by cross-tabulations (with classes for below, within, and above normal range). Laboratory data will be analyzed with respect to the normal ranges of values provided by the local laboratory and abnormal laboratory test results will be tabulated.

## **10.6 Vital Signs**

The statistical analysis of vital signs measurement will be mainly descriptive in nature.

## **10.7 Physical Examination**

Physical examination findings will be listed.

## **10.8 Adverse Events**

All original AEs terms will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). System organ classes and preferred terms will be used in the analyses.

Type and incidence of AEs, as well as severity and relatedness to the study medication will be tabulated. Special attention will be given to those patients who prematurely discontinue the study or the study medication due to an AE, or who experience a severe AE or an SAE.

## **10.9 Analysis of efficacy**

### **Primary endpoint:**

In order to assess <sup>99m</sup>Tc-rhAnnexin V-128 magnitude and dynamic range of uptake within areas affected by inflammation, SPECT/CT scans will be interpreted and graded by at least two independent experienced nuclear medicine physicians blinded from clinical data and other imaging modality results. Briefly, <sup>99m</sup>Tc-rhAnnexin V-128 uptake compared with background (e.g. physiological liver uptake) should be assessed for each affected area by nuclear medicine physicians using a 4-grade scoring system (e.g. 0, none; 1, mild or present but < to background uptake; 2, moderate or = to background uptake; 3, intense or > to background uptake). In case of discrepancies between the different readers, an adjudication process based on consensus should be put in place in order to obtain one final outcome for each area.

### **Secondary endpoints:**

Briefly, for quantifying the radioactivity uptake after injection of <sup>99m</sup>Tc-rhAnnexin V-128, ROIs will be drawn manually on the earliest images, and the shapes and sizes (i.e., number of pixels) will be kept constant over all subsequent images. Correction for background counts will be performed using an appropriate background region by subtracting the mean counts per pixel in the background region from the mean counts per pixel in each ROI to yield net counts. For each ROI, the geometric mean of net counts, corrected for physical decay, of total anterior and posterior counts will be calculated.

In order to compare SPECT/CT scans and MRI scans, diagnostic performances will be evaluated in terms of sensitivity (i.e., true positive rate), specificity (i.e., true negative rate), positive predictive value (i.e., proportion of positive values) and negative predictive value (i.e., proportion of negative values). To do this, physical examinations will be necessary to define sites of pain in patients with

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clinical suspicion or confirmed diagnosis of SpA. When sites of pain will match with abnormalities on diagnostic images, this will be regarded as true positive. McNemar test will be used to compare diagnostic performances of SPECT/CT and MRI techniques.

The determination of antibodies against rhAnnexin V-128 at baseline and post-treatment will be described as present or absent, and if applicable, its presence will be correlated as per the [REDACTED] objectives.

## **11 ADVERSE EVENTS AND OTHER SAFETY ASPECTS**

### ***11.1 Definition of Adverse Events***

An AE is defined as any untoward medical occurrence in a patient and which does not necessarily have a causal relationship with the study medication. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease, temporally associated with the use of a study medication, whether or not causally related to the study medication.

AEs will be reported from signing the informed consent onwards until the last study-related procedure. If the information of an untoward medical occurrence is collected before starting the intake of study medication, this information will be listed as a pre-treatment AE during statistical analysis.

### ***11.2 Definition of Serious Adverse Events***

A SAE is any untoward medical occurrence that at any dose:

- Results in death;
- Is life-threatening;  
Note: "life-threatening" refers to an event in which the patient is at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it was more severe;
- Results in persistent or significant disability/incapacity;
- Results in congenital anomaly or birth defect;
- Requires in-patient hospitalization or leads to prolongation of hospitalization.

Hospitalization or prolongation of hospitalization will not be considered as SAE in the following cases:

- Hospitalization planned before the patient inclusion,
- Hospitalization less than 24 h
- Hospitalization needed for the routine follow-up of the patient disease.

Medical and scientific judgment should be exercised in deciding whether expedited reporting is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require intervention to prevent one of the other outcomes listed in the definition above. These events, including those that may result in disability, should also usually be considered serious. Examples of

such events are intensive treatment in an emergency room or at home for severe allergic reactions that do not result in hospitalization.

The Investigator must report all SAEs by sending a completed SAE Reporting Form (Appendix III) to the Safety Officer designee within 24 h of becoming aware of the event.

All SAEs must be addressed to Advanced Accelerator Applications pharmacovigilance dept. with the following contact details:

E-mail: [pharmacovigilance@adacap.com](mailto:pharmacovigilance@adacap.com)

Fax: +33 450 993 634

When possible the SAE Form should be sent by e-mail.

### ***11.3 Investigator Reporting Requirements***

Throughout the study, the study staff will question the patient in a non-directive way as to the occurrence of AEs. The patient will also be instructed, when signing the informed consent, as from that moment, to contact the Investigator to report any study medication or non-study medication-related adverse or unusual event that occurs during participation to the study.

The study staff will record all these events in the patient's medical records and e-CRF, whether observed by the Investigator, the investigational staff, or spontaneously reported by the patient.

The Investigator will provide a complete description of the event in standard medical terminology, the date of onset and termination, severity, relationship to the study medication, action taken regarding the study medication, any treatment given, the outcome, and whether or not the event is considered as a SAE. If known, the Investigator should report the underlying illness or disorder rather than the individual signs and symptoms.

### ***11.4 Reporting of Serious Adverse Events***

In the case of a SAE, the Investigator must immediately (at the earliest possible time point within 24h of awareness) complete the SAE section of the e-CRF, reporting all information that is required by the Regulatory Authorities and contact the delegated Safety Officer designee, if needed. The name and contact details of the delegated Safety Officer designee will be available in the Investigator Site File (and will be updated when needed).

The minimum information required for immediate reporting is the event description, the patient ID, the study medication concerned, and the identifiable reporter (Investigator or designee). Even if not all the facts are known, an initial report should be made. The Investigator must provide follow-up information as soon as possible. If requested by the delegated Safety Officer designee, documents relevant to the diagnosis, treatment, and course of the event must be submitted (e.g. technical investigation reports, histology findings, hospital discharge documents). All documents must be anonymized with respect to the patient's personal identification data.

When the Investigator determines that there is not more information likely to be available, a final

report should be provided. The Sponsor or delegated CRO will assume responsibility for appropriate reporting of AEs to the regulatory authorities and the Independent Ethics Committee(s)/ Institutional Review Board(s) (IEC/IRB) according to local laws and regulations.

SAEs will be collected following the patient written consent to participate in the study. The collection of SAE information will continue to be reported by the Investigator for each patient until 30 days after the last study medication administration.

### ***11.5 Follow-up of Adverse Events***

All AEs occurring during the study are to be followed up until resolved or judged to be no longer clinically significant, or until they become chronic to the extent that they can be fully characterized. An assessment should be made at the last study-related visit for each patient. Certain long-term AEs cannot be followed until resolution within the settings of this protocol. In these cases follow-up will be the responsibility of the treating physician.

Since it is unpredictable how long such a follow-up might take, data from this follow-up generated after the patient's last study-related visit will be recorded by the Investigator. Full details regarding this follow-up will be described in the Clinical Study Report, if necessary.

If during AE follow-up the case has progressed to the level of "SAE", or if a new SAE whose relationship to the study medication could not be ruled out is observed, the situation must be reported immediately by the Investigator becoming aware of the information (considering that the "date of SAE onset" is the date of the first manifestations of that AE).

### ***11.6 Procedure in Case of Pregnancy***

Prior to clinical study enrolment, women of childbearing potential must be advised of the importance of avoiding pregnancy during clinical study participation and the potential risk factors for an unintentional pregnancy. The Investigator must report any pregnancy associated with investigational product exposure including conceptions occurring until 30 days after the last injection of radiotracer. The report should be carried out within 24 hours of pregnancy confirmation by sending a completed Pregnancy Reporting Form to the Safety Officer designee.

Appropriate pregnancy follow-up procedures should be considered if indicated. The Investigator must report follow-ups within 24 hours of the receipt of any new information on the course of the pregnancy, including perinatal and neonatal outcome, by sending a completed Pregnancy Reporting Form to the Safety Officer designee (Appendix IV).

## **12 TERMINATION OF THE STUDY**

Early termination of the study can occur in the following cases:

- When the visual review and analysis of the images of the first 5 patients by the Data Monitoring Committee does not enable to demonstrate the potential of the study product in terms of quality or efficacy, the Sponsor may discontinue the clinical study and send a written notice of the discontinuation along with the reasons to the investigator and applicable authorities.

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- When the Sponsor is aware of information on matters concerning the safety of the study product, as well as other important information that may affect proper conduct of the clinical study, the Sponsor may discontinue the clinical study and send a written notice of the discontinuation along with the reasons to the investigator and applicable authorities.
- If the investigator intends to discontinue participation in the study, the investigator must immediately inform the Sponsor of the discontinuation and the reason for it.
- The Sponsor reserves the right to discontinue the study at any time for failure to meet expected enrolment goals.

## **13 OPERATIONAL, ETHICAL, AND ADMINISTRATIVE CONSIDERATIONS**

### ***13.1 Study personnel***

All study staff will be informed of the study protocol through meeting and internal study initiation. Their task will be supported by internal SOPs specific to the study. The study nurse will be asked to collect blood and urine samples and to ship/send them to the appropriate laboratory.

### ***13.2 Data collection***

The study will be monitored by Advanced Accelerator Applications (AAA) according to the current SOP for the monitoring of studies. Shortly before the study starts, the Study Monitor will meet with the Investigator and Investigational Staff involved reviewing the procedures regarding study conduct and recording of data in the e-CRF. During the study, the Investigator will permit the Study Monitor to verify the progress of the study at the center as frequently as necessary. The Investigator will make the electronic data screens available, provide missing or corrected data, and sign the e-CRFs. Key data transcribed into the e-CRF will be reviewed against the source documents. Personal information will be treated as strictly confidential and will not be made publicly available. Any inconsistency between the source data and the data recorded in the e-CRF will be corrected.

The Sponsor will ensure that appropriate Quality Control (QC) steps are included into the different clinical processes to guarantee adequate protection of the study patients and quality of the data.

An independent Quality Assurance (QA) department, Regulatory Authorities and/or IECs/IRBs may review this study. This implies that auditors/inspectors have the right to inspect the study center(s) at any time during and/or after completion of the study and have access to source documents, including the patient's file. By participating in this study, the Investigator(s) agree(s) to this requirement.

For any data transfer, measures will be undertaken to protect patient data handed over against disclosure to unauthorized third parties and patient confidentiality will be maintained at all times.

### ***13.3 Data Review***

All data relating to the study must be recorded in the e-CRFs provided by the Sponsor. These e-CRFs should always reflect the latest observations on the patient's participation in the study.

Therefore, e-CRFs are to be completed within 5 days after the patient's visit. To avoid inter-observer variability, every effort should be made to ensure that the study determinations are completed by the same individual who made the initial ones at baseline. The Investigator must verify that all data entries in the e-CRFs are accurate and correct.

The monitor will review the e-CRFs and evaluate them for completeness and consistency. The e-CRF will be compared with the source documents to ensure that there are no discrepancies between critical data. All entries, corrections and alterations are to be made by the responsible Investigator or his/her designee. The monitor cannot enter data in the e-CRFs.

### **13.4 Data Clarification**

If corrections to an e-CRF are needed, the responsible monitor or data manager will raise a query. The appropriate investigational staff will answer queries.

### **13.5 Source Documents**

Source data must be available at the study center to document the existence of the study patients and substantiate the integrity of the study data collected. They must include the original documents related to the study, as well as the medical treatment and medical history documentation of the patient.

The source medical records should at least include the following information for each patient:

- Patient identification (name, date of birth, gender);
- Documentation of eligibility criteria, i.e. medical and medication history, physical examination;
- Participation in study (including study number);
- Study discussed, signed and dated ICF;
- Dates of all visits;
- Images/scans and reports;
- Documentation that protocol-specific procedures were performed;
- Study medication administration time and date;
- Receipt, dispensation and destruction of used/unused study medication;
- Record of all AEs and other safety parameters;
- Record of all previous and concomitant therapies;
- Date of study completion or reason for early discontinuation (if applicable).

The following documents are considered as source documents as well: nurse records and worksheets.

The author of an entry in the source documents must be identifiable. Direct access to source documentation (medical records) must be allowed for the purpose of verifying that the data recorded on the e-CRF are consistent with the original source data.

### **13.6 Clinical Study Monitoring**

Advanced Accelerator Applications or designee is responsible for periodic monitoring of the study to ensure that patients' rights, safety, and well-being are protected, that the study is properly conducted in adherence to the current protocol and ICH GCP, and that the data reported by the Investigator or designee are accurate, complete, and verifiable with the source documents. The assigned Clinical Study Monitor(s) will monitor the study in accordance with the monitoring guidelines. A copy of their Monitoring Log will be obtained at the study close-out visits.

### **13.7 Direct Access to Source Data/Documents**

The Investigator and the study center must provide all study-related records, as well as source documents in the instances when they are requested to by regulatory agencies including the local IRB of Cedars-Sinai Medical Center Office of Research of Compliance and FDA. The confidentiality of the patient's identity shall be well protected and consistent with local and national regulations when the source documents are patient to direct access.

### **13.8 Data Management**

Data management activities will be coordinated by the Sponsor with the support of a CRO as necessary.

All study-specific processes and definitions will be described in the Data Management Plan. Coding of AEs and Medical History terms will be performed using MedDRA; previous and concomitant medication will be coded using WHO codes.

### **13.9 Ethical Conduct of Clinical Study**

The Investigator(s) and all parties involved in this study should conduct the study in adherence to the ethical principles based on the Declaration of Helsinki, GCP, ICH guidelines, and the applicable laws and regulations.

ICH-GCP is an international ethical and scientific quality standard for designing, conducting, recording, and reporting study activities that involve the participation of human patients. Compliance with this standard provides public assurance that the rights, safety, and well-being of the patients are protected, consistent with the principles that have their origin in the Declaration of Helsinki, and that the study data are credible.

The Investigator and all study staff will conduct the study in compliance with the IEC/IRB approved version of this protocol. The protocol, ICF, any information provided to the patient, recruitment advertisements, and any amendments to these items will have IEC/IRB approval prior to their use in the study. Voluntary informed consent will be given by every patient in order to be screened for the study and prior to the initiation of any study-related procedures. The consent process must meet all applicable local laws. The rights, safety, and well-being of the patients are the most important considerations and prevail over the interests of science and society. All personnel

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involved in the conduct of this study must be qualified by education, training, and experience to perform their assigned responsibilities.

### **13.10 Authorities**

The protocol, name, and study center of the Investigators, the votes of the IEC(s)/IRB(s), as well as other locally required documents will be submitted to the regulatory authorities, according to local requirements for review and approval before the beginning of the study.

### **13.11 Patient Confidentiality**

Individual patient medical information obtained as a result of this study is considered confidential and disclosure to third parties is prohibited. Such information may only be given to a third party after approval of the patient, to the patient's general practitioner or to other appropriate medical personnel responsible for the patient's well-being.

The Sponsor, its board members, and its personnel shall not disclose any confidential information on patients obtained during the performance of their duties in the study without justifiable reasons. All individuals and organizations involved in conducting the study and/or processing the study data must pay very careful attention to protect the patients' privacy with appropriate measures, for example, by prohibiting the use of any private information that may identify a patient (e.g. name or address). These details shall be processed in accordance with the applicable local and regional laws.

### **13.12 Patient Information/Written Informed Consent**

According to ICH GCP (CPMP/ICH135/95) the patient must give consent to participate in the study, only after having been fully informed by the Investigator of the nature, significance, and implications of the study, as well as to the associated risks involved. Such meetings must be carried out on an individual basis, and adapted to the educational background and previous knowledge of the patient. Participation to this meeting will be documented in the patient's clinical chart. In the present protocol, patient will be informed by the rheumatologist about the scope and goals of the study based on the patient information sheet. Patient will have at least 24 hours to consider his participation to the protocol before signing the informed consent. Any study-related procedure will only take place after signed patient informed consent has been collected.

The patient will be instructed by the Investigator that the consent for study participation can be withdrawn at any time, without having to justify a reason, and that no disadvantageous consequences will follow regarding further medical treatment. The Investigator shall ask for the reason of premature termination without violating the patient's rights (ICH GCP Definition 4.3.4). Furthermore, the patient must be informed about insurance coverage and the corresponding patient obligations (see Patient Information). The ICF must be personally dated and signed by both the Investigator/Delegate and the patient. The patient receives one copy of the original patient information and Informed Consent Form signed and dated by both Investigator/Delegate and the patient.

The original of the ICF will be retained by the Investigator in the Investigator's File, who will confirm

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the patient's consent in the e-CRF. The patient will only be included in the study after written consent is given.

Furthermore, the Investigator is recommended to inform the patient's general practitioner of his/her participation in the study, provided that the patient has a general practitioner and the patient agrees to disclose this information. For this purpose, a copy of the patient information sheet and the patient's signed informed consent will be sent to the GP if the patient agrees with this and has ticked the appropriate tickbox on the Patient Information Consent Form.

### ***13.13 Arrangement for Use of Information and Publication of Clinical Study Data***

The present study will be registered on <http://clinicaltrials.gov/> and its results will be published.

All information regarding the investigational product under study in the outlined protocol and Advanced Accelerator Applications operations, such as patent applications, formulas, manufacturing processes, basic scientific data, or formulation information, supplied by AAA and not previously published, are considered confidential by AAA and shall remain the sole property of AAA. The Investigator agrees to use this information only to perform this study and will not use it for other purposes including publications and presentations without AAA's written consent.

It is understood by the Investigator that the information developed during the conduct of this study is considered confidential and will be used by AAA for the development of the specified investigational medication. This information may be disclosed as deemed necessary by AAA to other Investigators, other pharmaceutical companies, and to governmental agencies. To allow for the use of the information derived from this study and to ensure complete and thorough analysis, the Investigator is obligated to provide AAA with complete test results and all data developed in this study, and to provide direct access to source data/documents for study-related monitoring, audits, IEC/IRB review, and regulatory inspection.

Any publication or public presentation of the results of this study must be according to the Sponsor's standards. The first publication is coordinated by the Sponsor. The Investigator agrees that before he/she publishes any results of this study, he/she shall send the draft manuscripts and copies of the information to be presented to the Sponsor at least 30 working days before submission to a publisher or presentation. The Sponsor reserves the right to review these materials before submission for publication or presentation. This is not intended to restrict or hinder publication or presentation but instead to allow the Sponsor to protect proprietary information and to provide comments based on information that may not yet be available to the Investigator(s).

### ***13.14 Documents and Records Related to the Clinical Study***

The Investigator must retain e-CRFs and source documents of all enrolled patients (i.e. all patients who gave consent to be screened for the study), study medication disposition, and other documents required by regulation, in his/her possession or in an accessible area for at least 10

years after the completion of this study. The Investigator should take measures to prevent accidental or premature destruction of these documents. Under no circumstance shall the Investigator relocate or dispose any study documents before having obtained the Sponsor's written approval.

If it becomes necessary for the appropriate Regulatory Authority to review any documentation relating to this study, the Investigator must permit access to such reports.

Any difficulty in archiving and storage of clinical study documents must be discussed with the study monitor prior to the initiation of the study. The data and information collected during this study will be reported in Clinical Study Report(s) by the Sponsor.

### ***13.15 Sampling collection***

Biological samples collected at site for routine lab analysis (Hematology and Biochemistry) will be discarded after study analysis, according to local rules. For samples that are to be sent to a centralized foreign laboratory, i.e. for ELISA analysis, the Clinical Trial Agreement will be adapted according to local rules. These collected samples will be stored for up to 7 years (counting from when the last subject performed the last study visit) and then discarded, unless local rules, regulations or guidelines require different timeframes or different procedures, which will then be in line with the subject consent.

In case of patient consent withdrawal to the use of personnel data, biological samples will not be analyzed and will be destroyed. Samples collected will only be used for study purposes as per protocol.

### ***13.16 Protocol Amendment and/or Revision***

Any changes to the study, which arise after approval of the protocol, must be documented as protocol amendments and/or revisions. Depending on the nature of the amendment and/or revision, either IEC/IRB approval or notification is required. The changes will become effective only after the approval of the Sponsor, the regulatory authorities and the IEC/IRB (if applicable).

### ***13.17 Qualification of the Investigators***

The Investigator(s) should be qualified by education, language, training, and experience to assume responsibility for the proper conduct of the study. He/she should meet all qualifications specified by the applicable regulatory requirements and should provide evidence of such qualifications through an up-to-date curriculum vitae (Principal as well as all other Investigators) and/or other relevant documentation requested.

The Investigator should be thoroughly familiar with the appropriate use of the investigational product(s), as described in the protocol, the current Investigator's Brochure, the product information, and other information sources provided by the Sponsor.

The Investigator should be aware of, and should comply with, ICH-GCP and the applicable regulatory requirements. The Investigator should maintain a list of appropriately qualified persons to whom the Investigator has delegated significant study related duties.

### **13.18 Financial Disclosure**

The disclosed financial interest of the Investigator must be collected prior to enrolment of the first patient into the study, following study center completion, and 1 year following study completion. The Investigator should promptly update this information if any relevant changes occur during this period. Disclosable financial interests will be recorded on the Investigator Financial Disclosure Form, US FDA 1572 Form, and communicated to the regulatory authorities. Any Investigator(s) added as investigational staff must complete the Investigator Financial Disclosure Form (US FDA 1572 Form) at the beginning of his/her participation to the study. For any Investigator(s) leaving the site prior to study completion, an Investigator Financial Disclosure Form should be obtained at the end of his/her participation.

### **13.19 Insurance of Patients and Others**

The Sponsor has covered this study by means of insurance for the patient according to national requirements. The name and address of the relevant insurance company, the certificate of insurance, the policy number, and the sum insured are provided in the Investigator's File.

### **13.20 Investigator Indemnity**

The Sponsor shall be liable towards the patients in accordance with the provisions of the Clinical Study Law. Notwithstanding the foregoing; the Sponsor does not, however, agree to indemnify, defend, or hold the Investigator harmless against liability, damage, loss, cost or expense (including reasonable attorney's fees and expenses), including liabilities arising out of or in connection with claims of any nature by third parties, including, without limitation, in respect of bodily injury or death, arising out of or in connection with the negligence, wrongful acts or omissions or willful misconduct of the Investigator, the Institution, or its affiliates.

Including but not limited to:

- The making of unauthorized representations and warranties concerning the study medication or the study;
- The failure to obtain appropriate informed consent;
- The non-compliance with applicable rules or regulations;
- The failure to conduct the study in accordance with this protocol.

A condition of this indemnity obligation is that, whenever the Investigator has information from which it may be reasonably concluded that an incident of bodily injury, sickness, disease, or death has occurred, the Investigator must immediately notice the Sponsor of all pertinent data surrounding any such incident, and, in the event a claim is made or a suit is brought, the Investigator will assist the Sponsor and cooperate in gathering information with respect to the time, place, and circumstances, and in obtaining the names and addresses of the injured parties and available witnesses.

The Investigator shall not, except at his/her own cost, voluntarily make any payment or incur any expense in connection with any such claim or suit without the prior written consent of the Sponsor.

## 14 QUALITY ASSURANCE

The Sponsor is implementing and maintaining QA and QC systems with written SOPs to ensure that studies are conducted and data are generated, documented (record), and reported in compliance with the protocol, GCP, and applicable regulatory requirement(s).

The Sponsor or designee may arrange to inspect or audit the study center. The auditor is independent from the clinical monitoring and project management team at the Sponsor's site. The audit may include on-site review of regulatory documents, e-CRFs and source documents. The auditors will have direct access to these documents.

Medical imaging is the main research tool used in the present study. These tools are subject to strict quality control laws such as regular controls performed by the producer and regular internal controls.

## 15 REFERENCES

Benali K, Louedec L, Azzouna RB, Merceron O, Nassar P, Shoukr FA, Petiet A, Barbato D, Michel J-B, Sarda-Mantel L, Le Guludec D, Rouzet F. Preclinical validation of 99mTc–Annexin A5-128 in experimental autoimmune myocarditis and infective endocarditis: Comparison with 99mTc–HYNIC–Annexin A5. *Molecular Imaging*, 2014; 1–10.

Blankenberg FG, Katsikis PD, Tait JF, Davis D, Naumovski L, Ohtsuki K, Kojiwada S, Abrams MJ, Darkes M, Robbins RC, Maecker HT, Strauss HW. In vivo detection and imaging of phosphatidylserine expression during programmed cell death. *Proc. Natl. Acad. Sci. USA* 1998; 95: 6349–6354.

Blankenberg FG, Robbins RC, Stoot JH, Vriens PW, Berry GJ, Tait JF, Strauss HW. Radionuclide imaging of acute lung transplant rejection with annexin V. *Chest*, 2000; 117:834–840.

Blankenberg FG, Kalinyak J, Liu L, Koike M, Cheng D, Goris ML, Green A, Vanderheyden JL, Tong DC, Yenari MA. 99mTc–HYNICannexin V SPECT imaging of acute stroke and its response to neuroprotective therapy with anti-Fas ligand antibody. *Eur J Nucl Med Mol Imaging*, 2006; 33:566–574.

Blankenberg FG. Imaging the molecular signatures of apoptosis and injury with radiolabeled annexin V. *Proc Am Thorac Soc*, 2009; 6:469–476.

Bleil J, Maier R, Hempfing A, Schlichting U, Appel H, Sieper J, Syrbe U. Histomorphologic and histomorphometric characteristics of zygapophyseal joint remodeling in ankylosing spondylitis. *Arthritis Rheumatol*, 2014; 66(7):1745-54.

Dougados M, Baeten D. Spondyloarthritis. *Lancet*, 2011;377(9783):2127-37.

Ebringer A., T. Rashid, H. Tiwana, and C. Wilson, "A possible link between Crohn's disease and ankylosing spondylitis via Klebsiella infections," *Clinical Rheumatology*, vol. 26, no. 3, pp. 289–297, 2007.

Ehrenfeld M. Spondyloarthropathies. *Best Pract Res Clin Rheumatol*,2012; 26(1):135-45.

Engbersen R, Riksen NP, Mol MJ, Bravenboer B, Boerman O, Meijer P, Oyen WJG, Tack C, Rongen GA, Smits P. Improved resistance to ischemia and reperfusion, but impaired protection by

**Study Protocol**

ischemic preconditioning in patients with type 1 diabetes mellitus: a pilot study. *Cardiovascular Diabetol*, 2012; 11:124.

FDA Guidance for Industry and Researchers: The Radioactive Drug Research Committee: Human Research Without An Investigational New Drug Application, August 2010.

Geske FJ, Lieberman R, Strange R, Gerschenson LE. Early stages of p53-induced apoptosis are reversible. *Cell Death Differ*, 2001;8:182–191.

Hardy JW, Levashova Z, Schmidt TL, Contag CH, Blankenberg FG. 99mTc Annexin V-128 SPECT monitoring of splenic and disseminated listeriosis in mice: a model of imaging sepsis. *Mol Imaging Biol*. 2014 Nov 22. [Epub ahead of print]

Hammill AK, Uhr JW, Scheuermann RH. Annexin V staining due to loss of membrane asymmetry can be reversible and precede commitment to apoptotic death. *Exp Cell Res*, 1999;251:16–21.

Hermann KG, Bollow M. Magnetic resonance imaging of sacroiliitis in patients with spondyloarthritis: correlation with anatomy and histology. *Rofo*. 2014;186(3):230-7.

Hreggvidsdottir HS, Noordenbos T, Baeten DL. Inflammatory pathways in spondyloarthritis. *Mol Immunol*, 2014; 57:28-37.

Kietselaer BL, Narula J, Hofstra L. The Annexin code: revealing endocarditis. *Eur Heart J*, 2007; 28:948. 37.

Kenis H, Zandbergen HR, Hofstra L, Petrov AD, Dumont EA, Blankenberg FG, Haider N, Bitsch N, Gijbels M, Verjans JWH, Narula N, Narula J, Reutelingsperger CPM. Annexin A5 uptake in ischemic myocardium: demonstration of reversible phosphatidylserine externalization and feasibility of radionuclide imaging. *J Nucl Med*, 2010; 51:259–267.

Kietselaer BL, Reutelingsperger CP, Boersma HH, Heidendal GA, Liem IH, Crijns HJ, Narula J, Hofstra L. Noninvasive detection of programmed cell loss with 99mTc-labeled annexin A5 in heart failure. *J Nucl Med*, 2007; 48:562–567.

Kown MH, Strauss HW, Blankenberg FG, Berry GJ, Stafford-Cecil S, Tait JF, Goris ML, Robbins RC. In vivo imaging of acute cardiac rejection in human patients using 99mTechnetium labeled annexin V. *Am J Transplant*, 2001; 1:270–277.

Lorberboym M, Blankenberg FG, Sadeh M, Lampl Y. In vivo imaging of apoptosis in patients with acute stroke: correlation with blood-brain barrier permeability. *Brain Res*, 2006; 1103:13–19  
Lorberboym M, Feldbrin Z, Hendel D, Blankenberg FG, Schachter P. The Use of 99mTc-recombinant human annexin V imaging for differential diagnosis of aseptic loosening and low-grade infection in hip and knee prostheses. *J Nucl Med*, 2009; 50:534–537.

Mielants H., E. M. Veys, C. Cuvelier et al., “The evolution of spondyloarthropathies in relation to gut histology—II. Histological aspects,” *Journal of Rheumatology*, vol. 22, no. 12, pp. 2273–2278, 1995.

Narula J, Acio E, Narula N, Samuels LE, Fyfe B, Wood D, Fitzpatrick JM, Raghunath PN, Tomaszewski JE, Keely C, Steinmetz N, Green A, Tait JF, Leppo J, Blankenberg FG, Jain D, Strauss HW. Annexin-V imaging for noninvasive detection of cardiac allograft rejection. *Nature Medicine*, 2001; 7:1347- 1351.

Ogura Y, Krams SM, Martinez OM, Kapiwoda S, Higgins JP, Esquivel CO, Strauss HW, Tait JF, Blankenberg FG. Radiolabeled annexin V imaging: diagnosis of allograft rejection in an experimental rodent model of liver transplantation. *Radiology*, 2000; 214:795-800.

Ogura Y, Martinez OM, Villanueva JC, Tait JF, Strauss HW, Higgins JP, Tanaka K, Esquivel CO, Blankenberg FG, Krams SM. Apoptosis and allograft rejection in the absence of CD8+ T cells. *Transplantation*, 2001; 71:1827-34.

**Study Protocol**

Orchard T.R., H.Holt, L. Bradbury et al., "The prevalence, clinical features and association of HLA-B27 in sacroiliitis associated with established Crohn's disease," *Alimentary Pharmacology and Therapeutics*, vol. 29, no. 2, pp. 193–197, 2009.

Palm Ø., B. Moum, A. Ongre, and J. T. Gran, "Prevalence of ankylosing spondylitis and other spondyloarthropathies among patients with inflammatory bowel disease: a population study (the IBSEN study)," *Journal of Rheumatology*, vol. 29, no. 3, pp. 511–515, 2002

Peker C, Sarda-Mantel L, Loiseau P, Rouzet F, Nazneen L, Martet G, Vrigneaud JM, Meulemans A, Saumon G, Michel JB, et al. Imaging apoptosis with (99m)Tc-annexin-V in experimental subacute myocarditis. *J Nucl Med*, 2004; 45:1081–1086.

Poddubnyy D, Rudwaleit M. Early spondyloarthritis. *Rheum Dis Clin North Am*, 2012; 38(2):387-403.

Post AM, Katsikis PD, Tait JF, Geaghan SM, Strauss HW, Blankenberg FG. Imaging cell death with radiolabeled annexin V in an experimental model of rheumatoid arthritis. *J Nucl Med*, 2002; 43:1359–1365.

Rashid T. and A. Ebringer, "Gut-mediated and HLA-B27-associated arthritis: an emphasis on ankylosing spondylitis and Crohn's disease with a proposal for the use of new treatment," *Discovery Medicine*, vol. 12, no. 64, pp. 187–194, 2011.

Riksen NP, Oyen WJ, Ramakers BP, Van den Broek PH, Engbersen R, Boerman OC, Smits P, Rongen GA. Oral therapy with dipyridamole limits ischemia-reperfusion injury in humans. *Clin Pharmacol Ther*, 2005;78:52–59.

Riksen NP, Zhou Z, Oyen WJ, Jaspers R, Ramakers BP, Brouwer RM, Boerman OC, Steinmetz N, Smits P, Rongen GA. Caffeine prevents protection in two human models of ischemic preconditioning. *J Am Coll Cardiol*, 2006;48:700–707.

Rongen GA, Oyen WJ, Ramakers BP, Riksen NP, Boerman OC, Steinmetz N, Smits P. Annexin A5 scintigraphy of forearm as a novel in vivo model of skeletal muscle preconditioning in humans. *Circulation*, 2005; 111:173–178.

Rouzet F, Dominguez Hernandez M, Hervatin F, Sarda-Mantel L, Lefort A, Duval X, Louedec L, Fantin B, Le Guludec D, Michel JB. Technetium 99m-labeled annexin V scintigraphy of platelet activation in vegetations of experimental endocarditis. *Circulation*, 2008; 117:781–789.

Sarda-Mantel L, Michel JB, Rouzet F, Martet G, Louedec L, Vanderheyden JL, Hervatin F, Raguin O, Vrigneaud JM, Khaw BA, et al. (99m)Tc-annexin V and (111)In-antimyosin antibody uptake in experimental myocardial infarction in rats. *Eur J Nucl Med Mol Imaging*, 2006; 33:239–245.

Smith C, Gibson DF, Tait JF. Transmembrane voltage regulates binding of annexin V and lactadherin to cells with exposed phosphatidylserine. *BMC Biochemistry*, 2009; 10:5 doi:10.1186/1471-2091-10-5.

Steiman AJ, Pope JE, Thiessen-Philbrook H, Li L, Barnabe C, Kalache F, Kung T, Bessette L, Flanagan C, Haraoui B, Hochman J, Leclercq S, Mosher D, Thorne C, Bykerk V. Non-biologic disease-modifying antirheumatic drugs (DMARDs) improve pain in inflammatory arthritis (IA): a systematic literature review of randomized controlled trials. *Rheumatol Int*, 2013; 33:1105-1120.

Tahara N, Imaizumi T, Virmani R, Narula J. Clinical feasibility of molecular imaging of plaque inflammation in atherosclerosis. *J Nucl Med*, 2009; 50:331–334.

Tait JF, Smith C, Blankenberg FG. Structural requirements for in vivo detection of cell death with 99mTc-Annexin V. *J Nucl Med*, 2005; 46:807–815.

Tait JF, Smith C, Levashova Z, Patel B, Blankenberg FG, Vanderheyden J-L. Improved detection of

**Study Protocol**

cell death in vivo with annexin V radiolabeled by site-specific methods. *J Nucl Med*, 2006; 47:1546–1553.

Taki J, Higuchi T, Kawashima A, Fukuoka M, Kayano D, Tait JF, Matsunari I, Nakajima K, Kinuya S, Strauss HW. Effect of postconditioning on myocardial 99mTc-annexin-V uptake: comparison with ischemic preconditioning and caspase inhibitor treatment. *J Nucl Med*, 2007; 48:1301–1307.

Tang X-N, Wang Q, Koike M, Cheng D, Goris ML, Blankenberg FG, Yenari MA. Monitoring the protective effects of minocycline treatment with radiolabeled annexin V in an experimental model of acute focal cortical ischemia. *J Nucl Med*, 2007; 48:1822–1828.

Thimister PW, Hofstra L, Liem IH, Boersma HH, Kemerink G, Reutelingsperger CP, Heidendal GA. In vivo detection of cell death in the area at risk in acute myocardial infarction. *J Nucl Med*, 2003; 44: 391–396.

Tokita N, Hasegawa S, Maruyama K, Izumi T, Blankenberg FG, Tait JF, Strauss HW, Nishimura T. 99mTc-Hynic-annexin V imaging to evaluate inflammation and apoptosis in rats with autoimmune myocarditis. *Eur J Nucl Med*, 2003; 30:232–238.

Vriens PW, Blankenberg FG, Stoot JH, Ohtsuki K, Berry GJ, Tait JF, Strauss HW, Robbins RC. The use of technetium Tc99m Annexin V for the in vivo imaging of apoptosis during cardiac allograft rejection. *J Thorac Cardiovasc Surg*, 1998;116:844-53.

Weber U, Lambert RG, Ostergaard M, Hodler J, Pederson SJ, Maksymowych WP. Assessment of structural lesions in sacroiliac joints enhances diagnostic utility of magnetic resonance imaging in early spondylarthritis. *Arthritis Care Res*, 2010

Wending D, Claudepierre P. New bone formation in axial spondyloarthritis. *Joint Bone Spine*, 2013; 80(5):454-8.

Wright V., “Seronegative polyarthritis: a unified concept,” *Arthritis and Rheumatism*, vol. 21, no. 6, pp. 619–633, 1978.



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- 3) Place the vial in a suitable radiation shield appropriately labelled with date, time of preparation, volume and activity.
- 4) Aseptically add 2 mL  $\pm$  0.2 mL of Sodium Pertechnetate Tc-99m solution containing 740 MBq  $\pm$  74 MBq (20 mCi  $\pm$  2 mCi) to the vial in the lead shield (**Do not shake**).
- 5) Remove the vial from the lead shield and place it in an appropriately shielded roller. Leave the vial under slow rotation for 90 min.
- 6) Remove the vial from the shielded roller, inspect visually for the absence of particulate matter and discoloration and place it again in a lead shield.
- 7) Aseptically withdraw material using a sterile shielded syringe. The so-obtained solution is stable for 6 hours after completion of radiolabelling reaction. For the purpose of this study it is recommended to use it within 4 hours after preparation.
- 8) Perform the quality control according to the recommended methods in order to check the compliance to the specifications in table below:

*Table*  
*Specification of  $^{99m}\text{Tc}$  Annexin V-128 KIT product*

Test	Acceptance criteria	Method
Appearance	Clear solution, colorless and free from visible particles	Visual inspection
pH	5.0-5.8	pH indicator strips
Radiochemical purity (% $^{99m}\text{Tc}$ rhAnnexin V-128 species)	$\geq 90\%$	Thin layer chromatography (iTLC)
Radiochemical purity (% $^{99m}\text{Tc}$ Glucoheptonate)	$\leq 10\%$	Thin layer chromatography (iTLC)
Radiochemical purity (% $^{99m}\text{TcO}_2$ and $^{99m}\text{TcO}_4^-$ )	$\leq 8\%$	Thin layer chromatography (iTLC)

Cautionary Notes:

- Tc-99m pertechnetate eluate should be obtained from a generator which has been eluted within the last 24 h

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- Tc-99m pertechnetate eluate which is more than 6 hour old from the time of elution should NOT be used.

Stability evaluation was performed so far on the lab-scale, engineering and first GMP batches. Stability data indicate that the product is stable up to 12 months both at  $5^{\circ}\text{C} \pm 3^{\circ}\text{C}$  (long term condition) and up to 9 months at  $25^{\circ}\text{C} \pm 2^{\circ}\text{C}/60\% \text{ RH} \pm 5\% \text{ RH}$  (accelerated conditions), with high radiochemical purity (RCP, well above 90%) and good biological activity (Biopotency, higher than 90%). Since the Drug Product confirmed to be stable after 12 months of storage at the accelerated temperature conditions ( $25^{\circ}\text{C} \pm 2^{\circ}\text{C}/60\% \text{ RH} \pm 5\% \text{ RH}$ ), the current applied shelf-life is 12 months at  $5^{\circ}\text{C} \pm 3^{\circ}\text{C}$ .

The stability evaluation will be performed on future GMP batches in order to obtain complete stability data on at least three GMP batches of the Drug Product up to 12 months at the intended long term storage temperature ( $5^{\circ}\text{C} \pm 3^{\circ}\text{C}$ ). As mentioned above, the stability assessment is being also performed at the accelerated temperature condition ( $25^{\circ}\text{C} \pm 2^{\circ}\text{C}/60\% \text{ RH} \pm 5\% \text{ RH}$ ), up to 9 months.

**Stability of the Finished Product ( $^{99\text{m}}\text{Tc}$ -rhAnnexin V-128 radiolabelled Imaging Agent)**

The stability of the Finished Product is 6 h after labeling at room temperature. At this time the Radiochemical Purity is still  $\geq 90\%$ . For the purpose of this study it is recommended to inject the product within 4 h after completion of the radiolabelling reaction.

## Appendix II – SpA assessments

# BASDAI

## Bath Ankylosing Spondylitis Disease Activity Index



### Ankylosing Spondylitis International Federation

Please draw a mark on each line below to indicate your situation in the past 7 days:

Name: \_\_\_\_\_ Date: \_\_\_\_\_

<b>n</b>	<p><b>How would you describe the overall level of fatigue / tiredness you have experienced in the past week?</b></p> <p style="text-align: center;">             none <span style="margin-left: 20px;">0</span> <span style="margin-left: 20px;">1</span> <span style="margin-left: 20px;">2</span> <span style="margin-left: 20px;">3</span> <span style="margin-left: 20px;">4</span> <span style="margin-left: 20px;">5</span> <span style="margin-left: 20px;">6</span> <span style="margin-left: 20px;">7</span> <span style="margin-left: 20px;">8</span> <span style="margin-left: 20px;">9</span> <span style="margin-left: 20px;">10</span> <span style="margin-left: 20px;">very severe</span> </p>	<small>Evaluation by the doctor</small> <input style="width: 40px; height: 20px; border: 1px solid black;" type="text"/>
<b>o</b>	<p><b>How would you describe the overall level of AS neck, back or hip pain you have had in the past week?</b></p> <p style="text-align: center;">             none <span style="margin-left: 20px;">0</span> <span style="margin-left: 20px;">1</span> <span style="margin-left: 20px;">2</span> <span style="margin-left: 20px;">3</span> <span style="margin-left: 20px;">4</span> <span style="margin-left: 20px;">5</span> <span style="margin-left: 20px;">6</span> <span style="margin-left: 20px;">7</span> <span style="margin-left: 20px;">8</span> <span style="margin-left: 20px;">9</span> <span style="margin-left: 20px;">10</span> <span style="margin-left: 20px;">very severe</span> </p>	<input style="width: 40px; height: 20px; border: 1px solid black;" type="text"/>
<b>p</b>	<p><b>How would you describe the overall level of pain / swelling in joints other than neck, back or hips you have had in the past week?</b></p> <p style="text-align: center;">             none <span style="margin-left: 20px;">0</span> <span style="margin-left: 20px;">1</span> <span style="margin-left: 20px;">2</span> <span style="margin-left: 20px;">3</span> <span style="margin-left: 20px;">4</span> <span style="margin-left: 20px;">5</span> <span style="margin-left: 20px;">6</span> <span style="margin-left: 20px;">7</span> <span style="margin-left: 20px;">8</span> <span style="margin-left: 20px;">9</span> <span style="margin-left: 20px;">10</span> <span style="margin-left: 20px;">very severe</span> </p>	<input style="width: 40px; height: 20px; border: 1px solid black;" type="text"/>
<b>q</b>	<p><b>How would you describe the overall level of discomfort you have had in the past week from any areas tender to touch or pressure?</b></p> <p style="text-align: center;">             none <span style="margin-left: 20px;">0</span> <span style="margin-left: 20px;">1</span> <span style="margin-left: 20px;">2</span> <span style="margin-left: 20px;">3</span> <span style="margin-left: 20px;">4</span> <span style="margin-left: 20px;">5</span> <span style="margin-left: 20px;">6</span> <span style="margin-left: 20px;">7</span> <span style="margin-left: 20px;">8</span> <span style="margin-left: 20px;">9</span> <span style="margin-left: 20px;">10</span> <span style="margin-left: 20px;">very severe</span> </p>	<input style="width: 40px; height: 20px; border: 1px solid black;" type="text"/>
<b>r</b>	<p><b>How would you describe the overall level of morning stiffness you have had in the past week from the time you wake up?</b></p> <p style="text-align: center;">             none <span style="margin-left: 20px;">0</span> <span style="margin-left: 20px;">1</span> <span style="margin-left: 20px;">2</span> <span style="margin-left: 20px;">3</span> <span style="margin-left: 20px;">4</span> <span style="margin-left: 20px;">5</span> <span style="margin-left: 20px;">6</span> <span style="margin-left: 20px;">7</span> <span style="margin-left: 20px;">8</span> <span style="margin-left: 20px;">9</span> <span style="margin-left: 20px;">10</span> <span style="margin-left: 20px;">very severe</span> </p>	<input style="width: 40px; height: 20px; border: 1px solid black;" type="text"/>
<b>s</b>	<p><b>How long did your morning stiffness last from the time you wake up?</b></p> <p style="text-align: center;">             0 <span style="margin-left: 40px;">¼</span> <span style="margin-left: 40px;">½</span> <span style="margin-left: 40px;">¾</span> <span style="margin-left: 40px;">1</span> <span style="margin-left: 40px;">1¼</span> <span style="margin-left: 40px;">1½</span> <span style="margin-left: 40px;">1¾</span> <span style="margin-left: 40px;">2 hours or more</span> </p> <p style="text-align: center;"> <span style="margin-left: 20px;">(0)</span> <span style="margin-left: 20px;">(1)</span> <span style="margin-left: 20px;">(2)</span> <span style="margin-left: 20px;">(3)</span> <span style="margin-left: 20px;">(4)</span> <span style="margin-left: 20px;">(5)</span> <span style="margin-left: 20px;">(6)</span> <span style="margin-left: 20px;">(7)</span> <span style="margin-left: 20px;">(8)</span> <span style="margin-left: 20px;">(9)</span> <span style="margin-left: 20px;">(10)</span> </p>	<input style="width: 40px; height: 20px; border: 1px solid black;" type="text"/>

BASDAI =

BASDAI = (sum of questions 1 to 4 plus mean of questions 5 and 6) divided by 5



Name: \_\_\_\_\_

Date: \_\_\_\_\_

**Lateral lumbar flexion:** Patient stands with heels and buttocks touching the wall, knees straight, shoulders back, hands by the side. The patient is then asked to bend to the right side as far as possible without lifting the left foot/heel or flexing the right knee, and maintaining a straight posture with heels, buttocks, and shoulders against the wall. The distance from the third fingertip to the floor when patient bends to the side, is subtracted from the distance when patient stands upright. The manoeuvre is repeated on the left side.

Assessment:	> 20	18–20	15,9–18,9	13,8–15,8	11,7–13,79	9,6–11,6	7,5–9,5	5,4–7,4	3,3–5,3	1,2–3,2	< 1,2
Score:	0	1	2	3	4	5	6	7	8	9	10

mean of right/left

**Tragus-to-wall distance:** Maintain same starting position as above. Ensure head in as neutral position (anatomical alignment) as possible, chin drawn in as far as possible. Measure distance between tragus of the ear and wall on both sides, using a rigid ruler. Ensure no cervical extension, rotation, flexion or side flexion occurs.

Assessment:	< 10	10–12	13–15	16–18	19–21	22–24	25–27	28–30	31–33	34–36	> 36
Score:	0	1	2	3	4	5	6	7	8	9	10

mean of right/left

**Lumbar flexion (modified Schober):** With the patient standing upright, place a mark at the lumbosacral junction (at the level of the dimples of Venus on both sides). Further marks are placed 5 cm below and 10 cm above <sup>1</sup>. Measure the distraction of these two marks when the patient bends forward as far as possible, keeping the knees straight.

<sup>1</sup>) Among the "modified Schober"s published in the literature, the modification recommended by Macrae and Wright is used.

Assessment:	> 7,0	6,4–7,0	5,7–6,3	5,0–5,6	4,3–4,9	3,6–4,2	2,9–3,5	2,2–2,8	1,5–2,1	0,8–1,4	< 0,8
Score:	0	1	2	3	4	5	6	7	8	9	10

**Maximal intermalleolar distance:** Patient supine on the floor or a wide plinth, with the knees straight and the feet pointing straight up. Patient is asked to separate legs along the resting surface as far as possible. Distance between medial malleoli is measured.

Assessment:	> 119	110–119	100–109	90–99	80–89	70–79	60–69	50–59	40–49	30–39	< 30
Score:	0	1	2	3	4	5	6	7	8	9	10

**Cervical rotation:** Patient supine on plinth, head in neutral position, forehead horizontal (if necessary head on pillow or foam block to allow this, must be documented for future reassessments). Gravity goniometer placed centrally on the forehead. Patient rotates head as far as possible, keeping shoulders still, ensure no neck flexion or side flexion occurs.

> 85,0	76,6–85	68,1–76,5	59,6–68,0	51,1–59,5	42,6–51,0	34,1–42,5	25,6–34,0	17,1–25,5	8,6–17,0	< 8,6
0	1	2	3	4	5	6	7	8	9	10

mean of right/left

BASMI:  
(mean of 5 scores)

**\*) Remark:**

In the literature (Jenkinson et al: J Rheumatol 1994;21:1694–1698 and Jones et al: J Rheumatol 1995;22:1609) two different BASMI definitions have been published where the same measurement results lead to different BASMI values. The above is based on the newer definition of 1995 with scores 0 to 10 for each component.

Name: \_\_\_\_\_

Date: \_\_\_\_\_

Please draw a mark on each line below to indicate your level of ability with each of the following activities in the past 7 days:

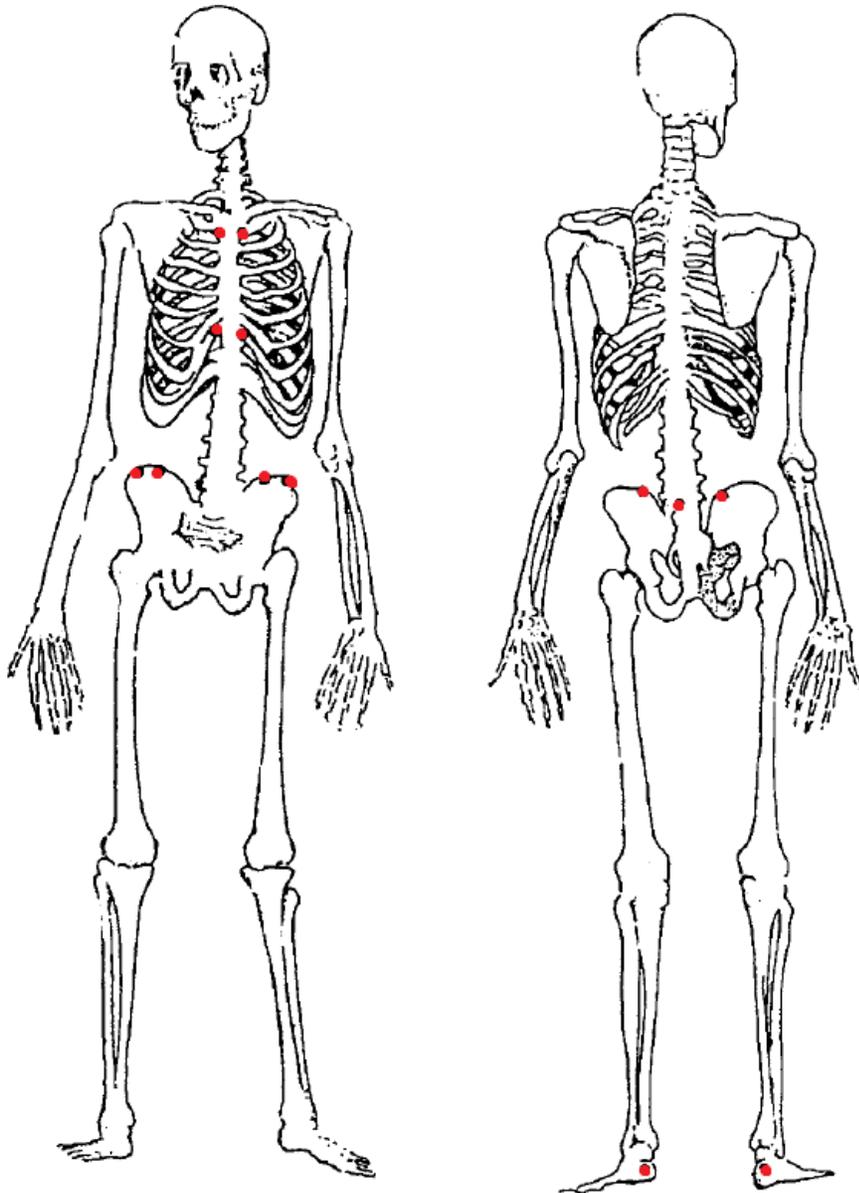


<b>n</b>	<b>Putting on your socks or tights without help or aids (e.g. sock aid)</b>		<input type="checkbox"/>
<b>o</b>	<b>Bending forward from the waist to pick up a pen from the floor without an aid</b>		<input type="checkbox"/>
<b>p</b>	<b>Reaching up to a high shelf without help or aids (e.g. helping hand)</b>		<input type="checkbox"/>
<b>q</b>	<b>Getting up out of an armless dining room chair without using your hands or any other help</b>		<input type="checkbox"/>
<b>r</b>	<b>Getting up off the floor without help from lying on your back</b>		<input type="checkbox"/>
<b>s</b>	<b>Standing unsupported for 10 minutes without discomfort</b>		<input type="checkbox"/>
<b>t</b>	<b>Climbing 12–15 steps without using a handrail or walking aid, one foot on each step</b>		<input type="checkbox"/>
<b>u</b>	<b>Looking over your shoulder without turning your body</b>		<input type="checkbox"/>
<b>v</b>	<b>Doing physically demanding activities (e.g. physiotherapy exercises, gardening or sports)</b>		<input type="checkbox"/>
<b>w</b>	<b>Doing a full day's activities whether it be at home or at work</b>		<input type="checkbox"/>

Evaluation by the doctor

BASFI =   
 (sum of answers 1 to 10 divided by 10)

## Maastricht Ankylosing Spondylitis Enthesis Score



MASES.

### Appendix III – SAE form

## Serious Adverse Event (SAE) Reporting Form

**ALL REPORTS MUST BE SIGNED AND DATED BY THE INVESTIGATOR.**

Please fax or send by e-mail the form to AAA Global Pharmacovigilance within 24 hours from awareness of event.

E-mail: [pharmacovigilance@adacap.com](mailto:pharmacovigilance@adacap.com);

Fax N°: +33 4 50 99 36 34;

Study Name	EudraCT N°	Center N°	Investigator N°	Country

**TYPE OF REPORT**

Initial

Completion of data

Follow-up

**1. PATIENT DATA - DO NOT SEND PATIENT IDENTIFIABLE DATA**

<b>Patient Initials</b> (first letter of First Name and first letter of Last Name): Last name <input type="text"/> First name <input type="text"/>	<b>Patient N°:</b>	<b>Sex:</b>	<b>Year of Birth</b> (yyyy):  <b>Age at onset of event:</b>	<b>Weight (kg):</b>  <b>Height (cm):</b>

**2. EVENT DETAILS**

<b>Date of onset</b> (dd/mm/yyyy):	<b>Diagnosis:</b>
<b>Description of SAE</b> (please state date of first use):	
<b>Seriousness Criteria</b> (check all that are relevant to the event): <input type="checkbox"/> Participant died <input type="checkbox"/> Hospitalisation or prolongation of existing hospitalisation <input type="checkbox"/> Life-threatening <input type="checkbox"/> Persistent or significant disability or incapacity	

<input type="checkbox"/> Congenital anomaly/ birth defect	<input type="checkbox"/> Other significant medical events	
<b>Severity of event:</b>	<input type="checkbox"/> Mild	<input type="checkbox"/> Moderate <input type="checkbox"/> Severe

3. STUDY TREATMENT							
Drug	Schedule	Route of administration	Start date (dd/mm/yyyy)	End date (dd/mm/yyyy)	Causally Related to Drug? <i>Tick either unrelated or possibly related</i>		Expected (Y/N)
					Unrelated	Possibly Related	
1.							
2.							
3.							
4.							

4. NIMPs (Non-investigational medicinal products)							
Are there any additional medications <b>used as part of the protocol?</b> <i>Such medications are referred to as NIMPs</i>							
Yes <input type="checkbox"/> No <input type="checkbox"/>							
<i>If yes, please complete the table below</i>							
NIMP(s)	Dose/schedule	Route of administration	Start date (dd/mm/yyyy)	End date (dd/mm/yyyy)	Causally Related to NIMP? <i>Tick either unrelated or possibly related</i>		Expected (Y/N/NA)
					Unrelated	Possibly Related	
1.							
2.							
3.							

5. CONCOMITANT DRUGS RELEVANT TO THE SAE <i>(do not include therapy used to treat the SAE)</i>							
<input type="checkbox"/> Tick box if no relevant concomitant medication							
Drug name	Dose/schedule	Route of administration	Reason for use	Start date (dd/mm/yyyy)	End date (dd/mm/yyyy)	Continued? (Y/N)	
1.							
2.							
3.							
4.							

5.						
6.						

**6. MEDICAL HISTORY (list relevant medical history):**  
 Tick box if no relevant medical history

Condition	Start Date (dd/mm/yyyy)	End date (dd/mm/yyyy)	Ongoing (Y/N)	Medication required Y/N
1.				
2.				
3.				
4.				

**7. RELEVANT TEST/LABORATORY FINDINGS** (include only the results relevant to the SAE diagnosis or course of SAE)

Test/lab finding	Unit	Date (dd/mm/yyyy)	Value	Reference range
1.				
2.				
3.				
4.				

Comment on test/laboratory findings (if none, mark as NA)

**8. ACTION TAKEN (check all that are relevant to the SAE)**

<input type="checkbox"/> No action taken	<input type="checkbox"/> Drug permanently discontinued due to this SAE	<input type="checkbox"/> Concomitant medication taken
<input type="checkbox"/> Drug schedule adjusted/temporarily interrupted	<input type="checkbox"/> Non-drug therapy given	<input type="checkbox"/> Hospitalisation/prolonged hospitalisation
<i>If multiple drugs used, please record which drug(s) have been adjusted/interrupted:</i>		

9. OUTCOME OF SAE		
<input type="checkbox"/> Completely recovered Date of recovery (dd/mm/yyyy)	<input type="checkbox"/> Condition still present and unchanged	<input type="checkbox"/> Recovered with sequelae Date (dd/mm/yyyy) Describe sequale:
<input type="checkbox"/> Condition deteriorated	<input type="checkbox"/> Condition improving	<input type="checkbox"/> Death Date of death (dd/mm/yyyy)
Autopsy: Yes <input type="checkbox"/> No <input type="checkbox"/> N/A <input type="checkbox"/> If Yes, include relevant information:		

10. ADDITIONAL INFORMATION

11. INFORMATION SOURCE	
Name, address and telephone number of PI	
Date of report (dd/mm/yyyy)	
PI signature	

**TO BE COMPLETED BY AAA GLOBAL PV (INTERNAL USE ONLY)**

DATE OF RECEIPT: (dd/mm/yyyy)	
AAA CASE TRACKING NUMBER:	
INFORMATION COMPLETE: <input type="checkbox"/> Yes <input type="checkbox"/> No	FOLLOW-UP REQUESTED: <input type="checkbox"/> YES <input type="checkbox"/> NO Details:
Name and signature:	

### PREGNANCY REPORTING FORM

**ALL REPORTS MUST BE SIGNED AND DATED BY THE INVESTIGATOR.**  
 Please fax or send by e-mail the form to AAA Global Pharmacovigilance within 24 hours from awareness of event.

E-mail: [pharmacovigilance@adacap.com](mailto:pharmacovigilance@adacap.com);  
 Fax N° : +33 4 50 99 36 34;

**N.B :** Fill the applicable fields and refer to Completion Guide of Pregnancy Reporting Form.

**TYPE OF REPORT:**     Initial     Completion of data     Follow-up n° .....

Study Name	EudraCT N°	Center N°	Investigator N°	Country

1. PATIENT INFORMATION			
<b>Patient initials</b> ( <i>first letter of Last Name and first letter of First Name</i> )  Last name <input type="text"/> <input type="text"/>  First name <input type="text"/> <input type="text"/>	<b>Patient N°:</b>	<b>Birthdate (YYYY):</b> <input type="text"/> <input type="text"/> <input type="text"/> <input type="text"/>	<b>Weight (kg):</b>  <b>Height (cm):</b>
		<b>Age:</b> .....	

2. PREGNANT WOMAN'S GENERAL MEDICAL HISTORY / BACKGROUND	
<b>BACKGROUND</b>  <b>Rhesus:</b> <input type="checkbox"/> Unknown <input type="checkbox"/> Rh - <input type="checkbox"/> Rh + <b>Smoking:</b> <input type="checkbox"/> Unknown <input type="checkbox"/> No <input type="checkbox"/> Yes ..... ( <i>pack-years</i> ) <b>Alcohol:</b> <input type="checkbox"/> Unknown <input type="checkbox"/> No <input type="checkbox"/> Yes ..... ( <i>glasses/day</i> ) <b>Drug abuse(s):</b> <input type="checkbox"/> Unknown <input type="checkbox"/> No <input type="checkbox"/> Yes ..... ( <i>detail</i> )	<b>PRIOR IMMUNIZATIONS</b>  <b>Rubella:</b> <input type="checkbox"/> Unknown <input type="checkbox"/> No <input type="checkbox"/> Yes <b>Toxoplasma:</b> <input type="checkbox"/> Unknown <input type="checkbox"/> No <input type="checkbox"/> Yes
<b>MEDICAL HISTORY</b>	<b>VIRAL SEROLOGY</b>

## Study Protocol

Hypertension:  Unknown  No  Yes  
 Diabetes mellitus:  Unknown  No  Yes  
 Epilepsy:  Unknown  No  Yes  
 Psychiatric diseases:  Unknown  No  Yes

HIV:  Unknown  No  Yes  
 Hepatitis:  Unknown  No  Yes

## 3. PREGNANT WOMAN'S GYNECOLOGICAL &amp; OBSTETRICAL HISTORY

## GYNECOLOGICAL HISTORY

Used contraception:  None  Oral  Local  Intra-uterine device  Other ..... (detail)  
 Regular menses:  No  Yes  
 Infertility treatment:  No  Yes ..... (detail)

## OBSTETRICAL HISTORY

Gravidity:  No  Yes ..... (detail)  
 Parity:  No  Yes ..... (detail)  
 In utero demise:  No  Yes ..... (detail)

Number of healthy live offspring: .....  
 Number of death offspring: .....  
 Number of malformed live offspring: .....

Abortions:  No  Yes

*If yes, please precise:*

Spontaneous ..... (detail)  
 Elective ..... (detail)  
 Therapeutic ..... (detail)

## 4. MATERNAL &amp; PATERNAL FAMILY HISTORY

Malformations:  Unknown  No  Yes ..... (detail)  
 Prematurely died children:  Unknown  No  Yes ..... (detail)  
 Psychomotor retardation:  Unknown  No  Yes ..... (detail)  
 Consanguinity:  Unknown  No  Yes ..... (detail)  
 Hereditary disease:  Unknown  No  Yes ..... (detail)  
 Other: ..... (detail)

## 5. CURRENT PREGNANCY STATUS AT TIME OF DETECTION

Last menstrual period: L J L J / L J L J / L J L J L J	Gestational age: .....	Polyzygotic pregnancy: <input type="checkbox"/> No <input type="checkbox"/> Yes
Estimated date of delivery: L J L J / L J L J / L J L J L J	Ultrasound-estimated gestational age: .....	Ectopic pregnancy: <input type="checkbox"/> No <input type="checkbox"/> Yes

## 6. COURSE OF CURRENT PREGNANCY

## DELETERIOUS EXPOSURES DURING PREGNANCY

## PATHOLOGY (IES) DURING PREGNANCY



## Study Protocol

<b>Gestational age:</b> ..... (weeks)	<input type="checkbox"/> Induced delivery <input type="checkbox"/> Caesarean section	<b>Normal placenta:</b> <input type="checkbox"/> No <input type="checkbox"/> Yes <input type="checkbox"/> Unknown	<b>Amniotic fluid:</b> <input type="checkbox"/> Clear <input type="checkbox"/> Turbid
<b>Postpartum maternal condition:</b>	<input type="checkbox"/> Normal <input type="checkbox"/> Abnormal	..... (detail)	
<b>Intrapartum medication:</b>	<input type="checkbox"/> Unknown <input type="checkbox"/> No <input type="checkbox"/> Yes	..... (if yes, detail)	

## 9. NEWBORN

<b>Sex:</b> <input type="checkbox"/> F <input type="checkbox"/> M	<b>Preterm:</b> <input type="checkbox"/> No <input type="checkbox"/> Yes	<b>Intensive care:</b> <input type="checkbox"/> No <input type="checkbox"/> Yes <input type="checkbox"/> Unknown
<b>Weight:</b> ..... (unit)	<b>Dysmature:</b> <input type="checkbox"/> No <input type="checkbox"/> Yes	<b>Malformation:</b> <input type="checkbox"/> No <input type="checkbox"/> Yes ..... (detail)
<b>Length:</b> ..... (unit)	<b>APGAR score:</b> ..... (1 min) ..... (5 min)	<b>Neonatal pathology:</b> <input type="checkbox"/> No <input type="checkbox"/> Yes ..... (detail)
<b>Cranial perimeter:</b> ..... (unit)		<b>Breastfeeding:</b> <input type="checkbox"/> No <input type="checkbox"/> Yes
<b>Transfer to NICU / pediatrics:</b> <input type="checkbox"/> No <input type="checkbox"/> Yes ..... (duration)	<b>Department address:</b> .....	
<b>Immediate outcome:</b> .....	<b>Child's follow-up performed by:</b> .....	

## 10. INITIAL REPORTER

Occupation:	<b><u>If the reporter is a patient or not a Healthcare Professional:</u></b> Has the patient given the authorization to AAA to follow up the pregnancy with its treating Doctor? <input type="checkbox"/> Yes <input type="checkbox"/> No
Full name:	
Organization/Address:	Date & signature _____
Telephone:	<b><u>Details of the treating Doctor (if different from the reporter):</u></b>
Fax:	Occupation:
Email:	Full name:
Date & signature _____	Organization/Address:
	Telephone/Fax:
	Email:

## ADMINISTRATIVE INFORMATION (for internal use only)

<b>AAA Case #:</b> .....	<b><u>Initial reception by:</u></b> NAME: OCCUPATION: LOCAL AFFILIATE/COUNTRY:
<b>TYPE DE RAPPORT:</b> <input type="checkbox"/> Initial <input type="checkbox"/> Follow up n°: .....	
Reception date by AAA PV department L L L / L L L / L L L L L L	

Reception date by a partner [ ]/[ ]/[ ]	DATE: [ ]/[ ]/[ ]
	SIGNATURE _____

*Data collected during a Pharmacovigilance investigation may be stored in a database according to Regulation (UE) no 520/2012, Regulation (CE) no 726/2004 and directive 2001/83/CE.*