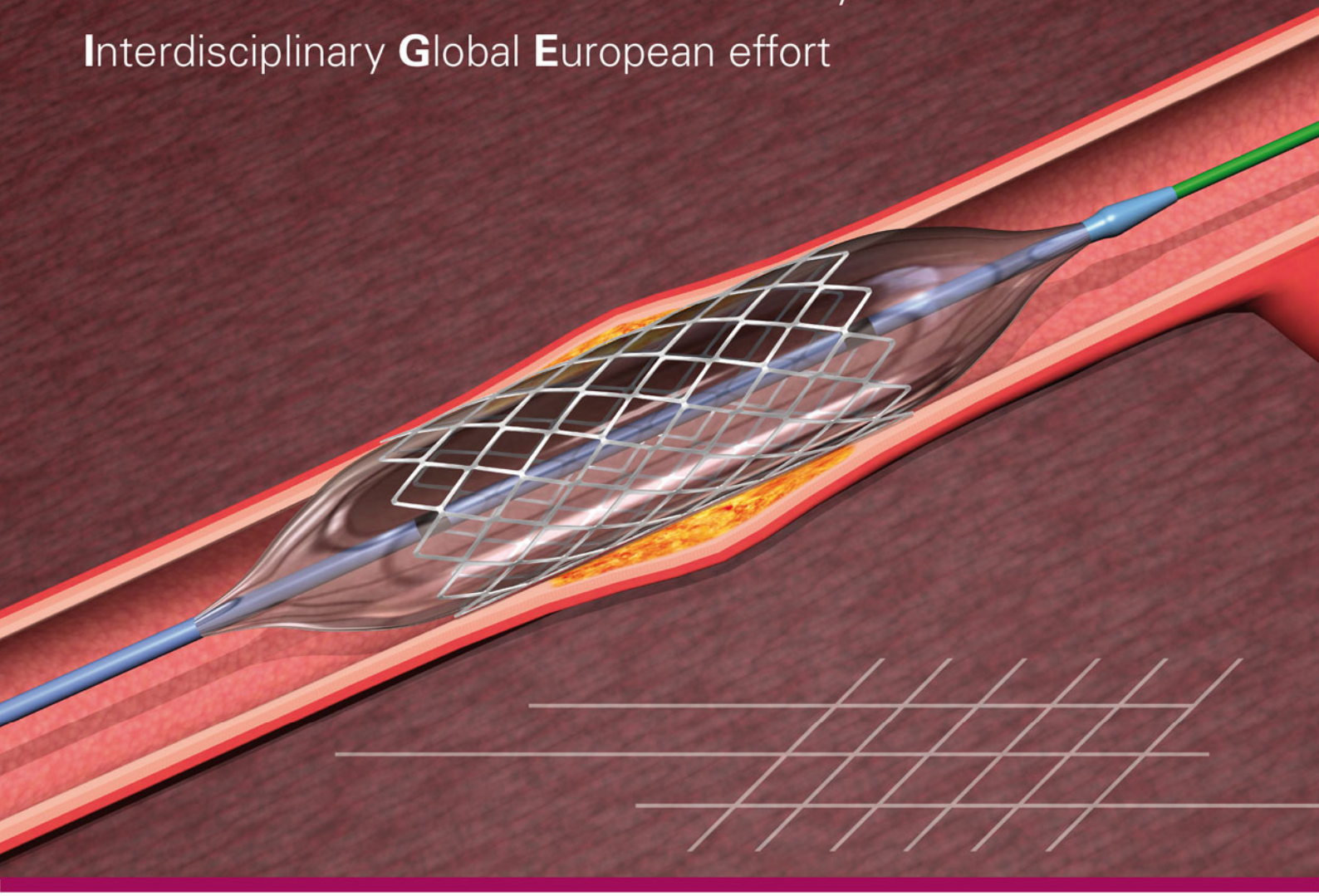
PRESTIGE

PREvention of late **S**tent **T**hrombosis by an
Interdisciplinary **G**lobal **E**uropean effort



SEVENTH FRAMEWORK PROGRAMME

Grant Agreement number:

260309

Project acronym:

PRESTIGE

Project title:

PREvention of late Stent Thrombosis
by an Interdisciplinary Global European effort

Summary

Regarding the Activities of the PRESTIGE Consortium

from 1 December 2010 to 31 May 2012

1. Summary description of project context and objectives

The development of drug-eluting stent (DES) therapy is a significant milestone in the care of patients with obstructive coronary artery disease. In particular DES therapy represented a significant gain in the battle against coronary restenosis, the major limitation of percutaneous coronary intervention (PCI) in the bare metal stents (BMS) era. With the passage of time however it has become clear that this advancement has come at the cost of a small excess of thrombotic stent occlusion – stent thrombosis (ST) – particularly late after DES implantation.

The principal objectives of PRESTIGE may be considered under 3 interrelated categories. Firstly, the principal medical objective is to develop new strategies to prevent late ST at a cost of minimum bleeding risk. Secondly, the principal scientific objective is to dissect the mechanisms contributing to the occurrence of late ST. A better mechanistic understanding of late ST is a conditio sine qua non for the development of more specific anti-thrombotic regimens that have minimal effects on normal haemostasis. Thirdly, the major technological objective of PRESTIGE is to develop novel imaging technologies allowing for early diagnosis and a better risk prediction of late ST as well as to evaluate optimized stent designs that promote enhanced vascular healing.

The scientific work of PRESTIGE is divided into 4 interrelated work packages (WP) (see **Figure 1**):

WP1 is focused on better mechanistic understanding of the molecular and cellular events triggering late ST. In dissecting the basic pathophysiology leading to late ST, PRESTIGE has generated a collaborative platform integrating the expertise of European centres of excellence in platelet thrombosis, endothelial biology and coagulation aimed at unravelling the early steps initiating the ST cascade as well as characterizing the key similarities and differences of the processes involved in late ST as compared with normal haemostasis.

WP2 is aimed at developing and validating novel strategies to reduce late ST: Using a multidisciplinary translational approach combining basic science, preclinical

research and small- and medium-sized enterprises as well as the mechanistic insights gained in WP1, this WP is focused on the realisation of novel stent devices with reduced thrombotic risk and improved healing characteristics.

WP3 will deliver and evaluate the role of novel imaging technologies for the assessment of vascular healing and dysfunction after coronary stenting: Utilizing a multidisciplinary approach bringing together imaging engineers basic researchers and clinical imaging specialists this WP is focused on twin imaging modalities of optical coherence tomography (OCT) and near-infrared fluorescence molecular imaging (NIRF). The aim is the thorough validation of imaging parameters mitigating an increased risk of ST via tissue characterization analysis, as well as applied histopathological correlation studies in preclinical models, autopsy specimens and in man. The ultimate clinical goal is the application of imaging technology towards the individualization of prolonged anti-platelet therapy and the consequent amelioration of global bleeding risk.

WP4 is performing multi-stranded characterisation of patients with late ST: The specific objective of PRESTIGE WP4 was the establishment of a pan-European stent thrombosis registry encompassing a collaborative network of centres from Central, Southern, Eastern and North-western Europe. Using a multi-centre case-control model with a recruitment phase of 3 years the PRESTIGE Registry is recruiting at least 500 patients presenting with late ST as well as 1,000 matched control patients. All patients with ST recruited in PRESTIGE undergo a multi-stranded analysis, including an in-depth description of patient-demographic and procedure-related factors, analysis of genetic and bio-markers, platelet function testing, histopathological analyses of the thrombus retrieved from the involved coronary artery and intracoronary imaging of the involved segment of the coronary artery using OCT and intravascular ultrasound. An optimized identification of patients at risk for ST is prerequisite for the implementation of individualized anti-thrombotic therapies.

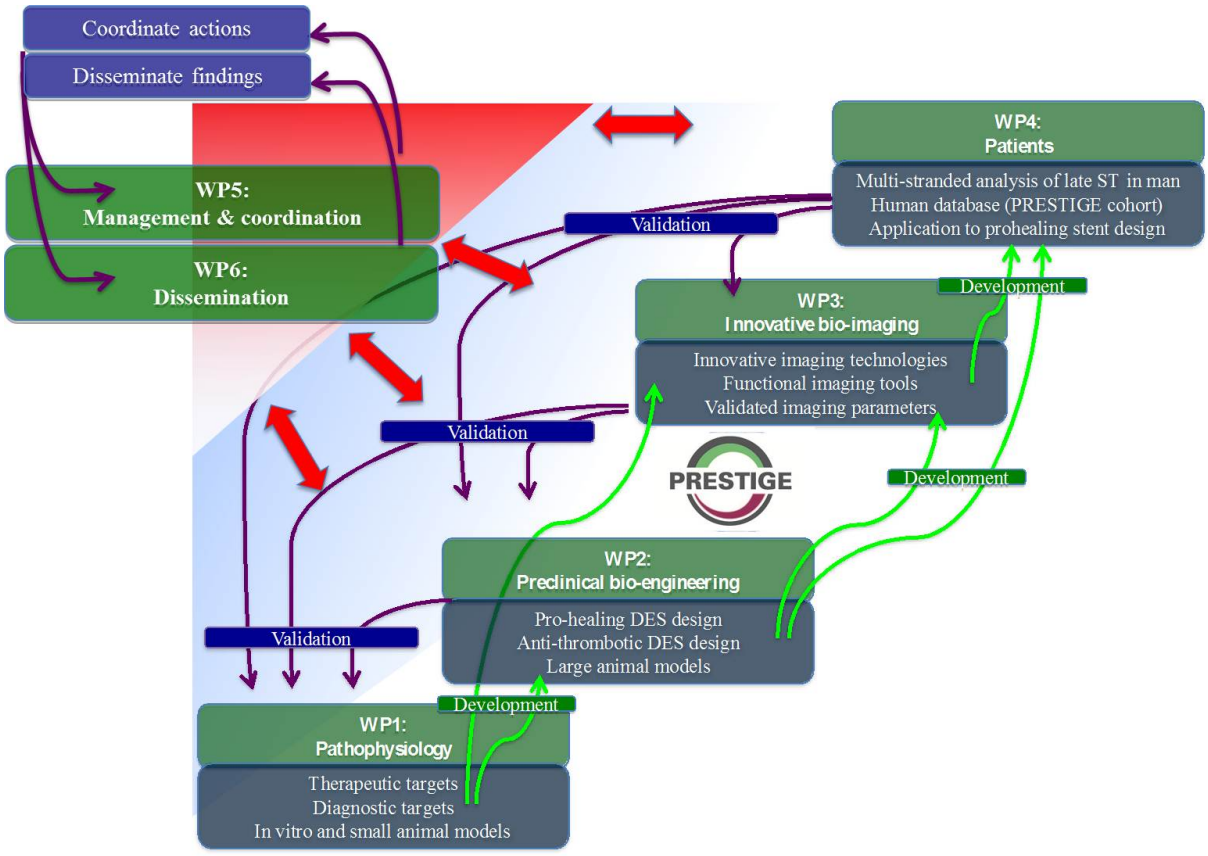


Figure 1. Overall strategy of PRESTIGE

2. Description of the work performed since the beginning of the project and the main results achieved so far

WP1

BIO together with DHM has developed and tested a mouse-dedicated stent platform for abdominal aortic implantation requiring a nominal diameter of 1.1-1.2 mm. This stent is mounted on a novel stent delivery system, which constitutes the second major component of the mouse stenting system (see **Figure 2**). INSERM U698/U949 have dissected the molecular and cellular mechanisms of stent thrombosis using a newly established model of cobalt-chromium (CoCr) disks provided by BIO. CoCr discs recruit platelets at their surface in a highly efficient manner. Molecular determinants of platelet adhesion and activation on CoCr discs were studied with the use of function-blocking antibodies and pharmacological inhibitors that showed that the GPIb-IX complex is essential for platelet attachment to the CoCr discs in vitro. The use of transgenic mouse platelets confirmed the importance of the GPIb-V-IX complex. Moreover we showed that activation of the platelet ADP receptor P2Y12 and of the platelet collagen receptor GPVI contribute to the procoagulant response of

platelets during interaction with CoCr. We also found that CoCr also directly activates coagulation independent from platelets. In addition we showed that neutrophils release extracellular traps, which trigger activation of the intrinsic pathway of coagulation in vitro (DHM).

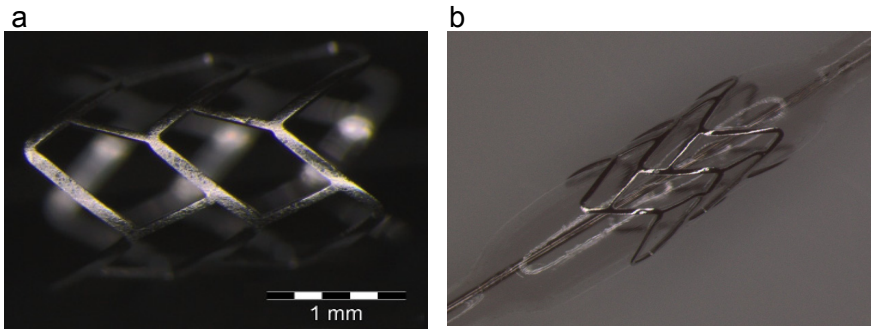


Figure 2. Mouse stent deployment. (a) mouse dedicated stent of 2.5 mm length; (b) mouse stent on first generation delivery device

WP2

DHM and BIO coupled RGD peptidomimetics onto stent surfaces to attract endothelial cells. RGD peptidomimetics were shown to bind to amino-functionalized stent surfaces, which resulted in increased adhesion of endothelial cells (**Figure 3**). CoCr discs were modified with aminosilane or poly-urethane polymers to provide a substrate for the coupling of novel aptamers attracting endothelial progenitor cells. Aminosilanization increased endothelial cell adhesion to CoCr discs. Cell adhesion assays were established for endothelial cells under both static and dynamic conditions. RTU, KIZ, BIO and NEO established novel coating technologies based on different monolayer/multilayer chitosan polymers. Plasma treatment of these polymeric coatings resulted in functionalization by creating reactive aminogroups. DHM, BIO, INSERM and ULEIC established rabbit and pig models of stent implantation. INSERM showed that novel dextran-based copolymeric stent coatings decreased platelet adhesion in vitro. Sub-contractor CNR examined poly-urethane and aminosilane polymers for their potential to trigger inflammation. DHM completed an animal study with completely bioabsorbable magnesium showing excellent biocompatibility and absence of severe inflammation after complete resorption of the devices.

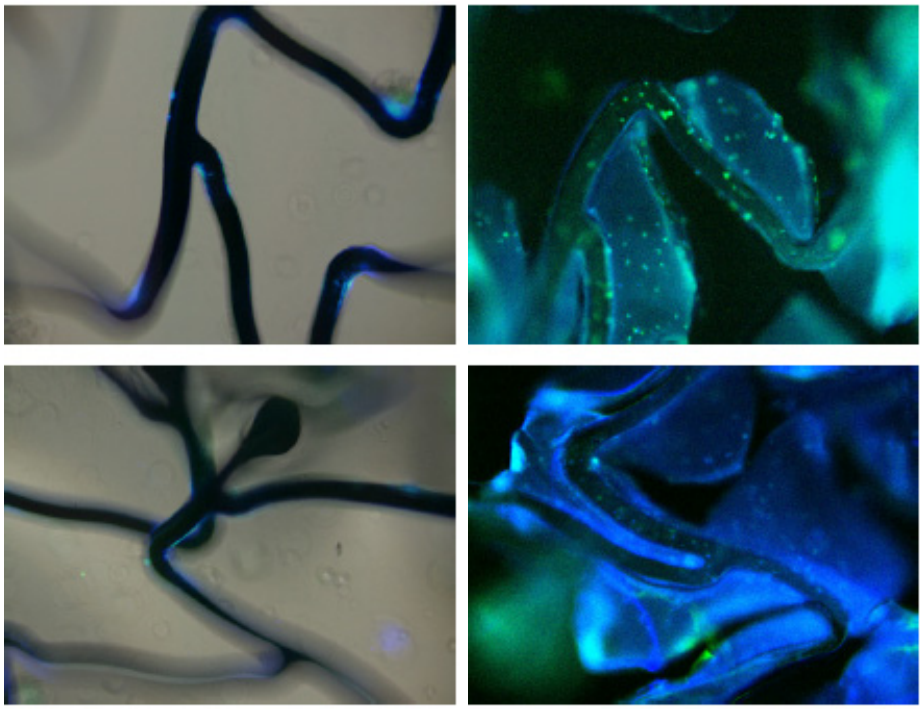


Figure 3. Endothelial cells on uncoated and RGD-coated drug eluting stents. Representative en face views of uncoated (left panels) and RGD-coated (right panels) drug-eluting stents. Endothelial cells have been labelled with phalloidin-488 to stain for cytoskeletal acting filaments. Endothelial cells appear as green fluorescent dots. Note, there is greater adhesion of endothelial cells after RGD coating compared to the uncoated control drug eluting stent.

WP3

Stented arteries derived from atherosclerotic rabbit models and human ex vivo specimens were imaged by OCT and examined histopathologically. Fluorescent-tagged antibodies were used to delineate endothelial regrowth after arterial denudation using OCT. A dedicated preclinical animal study was conducted by DHM, BER and HMGU to correlate intravascular OCT imaging with histopathology. Innovative tissue characterization software was used to distinguish mature from immature neointimal tissue. A novel NIRF imaging catheter was constructed to enable two-dimensional intravascular in vivo imaging of fluorescent probes in arteries with similar dimensions to those of human coronary arteries. Molecular targets with distinct biological processes of interest were labelled by fluorescent probes and imaged by the NIRF system using an atherosclerotic animal model. An animal model of non-occlusive stent thrombosis was developed to evaluate a specific radiotracer with high affinity for activated platelets and for detection of in-stent thrombi using scintigraphy. Finally human coronary arteries were examined for OCT-histopathology correlation.

WP4

An OPENCLINICA database has been installed at K.U.LEUVEN as an e-CRF application, used for electronic data capture and clinical data management; 117 ST-cases have been acquired. A DNA bank of patients with stent thrombosis has been established in DHM, NIE and ULEIC; a core lab for assessment of platelet function in patients with stent thrombosis has been established in NIE. Centralized histopathological analysis of thrombi is established in DHM; 75 samples were analysed. Core labs for analysis of OCT were established by DHM, BER and K.U.LEUVEN. As of 31.05.2012, 50 OCT examinations, performed in patients with stent thrombosis have been collected for analysis. DHM is already well advanced in recruiting of a cohort of patients with OCT follow-up in patients treated with enhanced biocompatibility biodegradable polymer and polymer-free DES platforms.

3. Expected final results and their potential impact and use

The overall impact of our project on the reduction of in-stent thrombosis is expected to result from both the scientific knowledge acquired and the new technologies realised over the course of the project. This will be accomplished through consecutive translational research steps within a consortium combining experienced investigators with proven track-records in the scientific fields relevant to this topic.

The impact on prediction of late stent thrombosis

At a patient level, a sophisticated risk stratification model including genetics, platelet function testing and bioimaging techniques will provide a basis for a comprehensive predictive approach to the problem of ST. With the elucidation of *in vivo* parameters associated with stent thrombosis, a pre-requisite for proper prediction will be met. Clinically relevant surrogate parameters will be established in preclinical and human autopsy studies and transferred to the clinical investigation of patients receiving DES. The unique comprehensive evaluation of preclinical and clinical bioimaging parameters against the gold standard of histopathology in stented human coronary arteries will ensure a reliable validation of surrogate markers of delayed arterial healing which is critical for the accurate prediction of late ST.

The impact on prevention of late stent thrombosis

All work packages of the current project are designed to provide novel insights into the mechanistic understanding of stent thrombosis, thereby generating a basis for innovative preventive strategies. Prevention of stent thrombosis is of utmost interest to the consortium owing to its high case mortality rate. It will be achieved through design and establishment of novel anti-thrombotic coatings for stent devices that will be based on dedicated research in platelet biology and coagulation pathways. Further, novel biodegradable stent components will be tested for their ability to improve vascular healing and decrease the potential for late ST after implantation. All preclinical and clinical investigations are designed to provide a maximum support for clinical competitiveness and technical innovation.

The role of PRESTIGE clinical trials and registries

The conduct of clinical patient trials and the implementation of the pan-European PRESTIGE stent thrombosis registry will enable the consortium to gather highly exploitable information relevant to the understanding of different DES types in different vascular lesions, which will ultimately provide the background for the establishment of novel strategies to prevent ST.

Genetics: the added value of a European project

The identification of genetic factors underlying the occurrence of late ST could be a major breakthrough in cardiovascular research, likely resulting in new therapeutic modalities. Due to differing genetic background across European countries, the current consortium provides an ideal platform for a highly exploitable genetic analysis.

The social impact

Addressing concerns regarding the late safety of drug-eluting stents is vital to the further widespread adoption of these devices. The impact of drug-eluting stents on enhancing quality of life, reducing patient morbidity (and possibly mortality), and reducing economic costs is very sizeable and optimizing DES utilization and reducing the burden of ST will result in significant societal benefit.

The impact on European cardiovascular research networks

Meeting the challenges involved in reducing the burden of ST is best accomplished utilizing a pan-European approach, drawing on the proximity and interrelationship of numerous excellent researchers in the respective fields of medicine and industry. A European approach not only facilitates the interaction of all consortium partners but also provides a broad spectrum of outstanding research expertise drawn from a range of different European countries. The establishment of such networks between researchers and industrial partners may also provide great future potential and flexibility to meet the changing needs of clinical research in interventional cardiology. We envisage that our investigations have the potential to strengthen the European pre-eminence in cardiovascular research.

The impact on other vascular diseases

Additional impact is expected on several other vascular disease entities including ***de novo coronary artery disease***, percutaneous intervention for **c*erebrovascular disease*** and ***peripheral artery disease***.

4. Address of the project public website

Further details of the PRESTIGE project are available at a public-access website (**Figure 4**) at www.prestige-fp7.eu

Figure 4. PRESTIGE project home page