

Aberrant endothelial and smooth muscle progenitor cell frequency and function in Type 2 diabetes: a predisposing factor for increased cardiovascular risk?

Published: 13-02-2009

Last updated: 06-05-2024

1) Determine differences in EPC and SMPC availability in the peripheral blood of T2DM patients with or without coronary artery disease (CAD) or peripheral artery disease (PAD) as well as in age- and sex-matched non-diabetic subjects with or without...

Ethical review	Approved WMO
Status	Recruitment stopped
Health condition type	Coronary artery disorders
Study type	Observational non invasive

Summary

ID

NL-OMON35549

Source

ToetsingOnline

Brief title

Vascular progenitor cells in Type 2 diabetes

Condition

- Coronary artery disorders
- Diabetic complications
- Arteriosclerosis, stenosis, vascular insufficiency and necrosis

Synonym

atherosclerosis, cardiovascular disease

Research involving

Human

Sponsors and support

Primary sponsor: Universitair Medisch Centrum Groningen

Source(s) of monetary or material Support: Diabetes Fonds

Intervention

Keyword: atherosclerosis, cardiovascular disease, progenitor cells, Type 2 diabetes

Outcome measures

Primary outcome

From the peripheral blood obtained PBMCs will be isolated using Ficoll density gradient centrifugation. Isolated cells will then be subjected to:

1) flowcytometric analyses:

These phenotypic analyses allows quantification circulating endothelial and smooth muscle progenitor cells (EPCs and SMPCs, respectively) in the peripheral blood of diabetic and non-diabetic subjects and correlate frequencies of these specific cell subsets to the presence of cardiovascular disease.

2) flowcytometric cell sorting:

These analyses allow purification of specific EPC and SMPC subsets which will be used for more in-depth functional and molecular analyses as described below.

3) in vitro EPC and SMPC culture:

These analyses allow functional studies on EPCs and SMPCs like tube

formation, anti-thrombotic activity, differentiation

into mature ECs and SMCs, SMC contractility, and matrix production.

4) molecular analyses:

In order to gain insight into the molecular pathways involved in aberrant

EPC and SMPC function as determined under

point 3, mRNA and proteins will be isolated from purified cells subsets

and/or after expansion/differentiation in vitro.

Isolated mRNA and proteins will then be subjected to gene expression

analysis (qRT-PCR and microarray analysis

[Illumina platform] and protein expression analysis (Western-blotting and

kinome profiling [PepChip platform]),

respectively.

5) histological analyses

From a subpopulation of patients with moderate to severe peripheral artery

disease the endarterectomy specimen will be collected and

processed for histological and molecular biological analyses. Results of

these analyses will be correlated with the in vitro generated data on

cultured cells using the methods described above (1 to 4).

Collectively, the results of these analyses will contribute to the

determination of the level of vascular progenitor cell dysfunction in T2D and

to the identification of novel molecular targets for therapeutic intervention aiming at modulating vascular progenitor cell frequency and function in order to attenuate development of cardiovascular disease and restenosis in T2DM.

Secondary outcome

n.a.

Study description

Background summary

Individuals with Type 2 diabetes mellitus (T2DM) have increased rates and severity of cardiovascular disease (peripheral artery disease [PAD] and coronary artery disease [CAD]) and restenosis. The pathogenetic mechanism(s) underlying increased cardiovascular disease and restenosis in T2DM is largely unknown and adequate treatment strategies are lacking. We hypothesize that a disturbed balance in vascular progenitor cells involved in maintenance of endothelial integrity (i.e. endothelial progenitor cells [EPCs]), and atherogenesis and neointima formation (i.e. smooth muscle progenitor cells [SMPCs]) is responsible for increased susceptibility for cardiovascular disease and restenosis in T2DM. Mechanistic understanding of the molecular pathways involved in aberrant vascular progenitor cell function in T2DM is anticipated to be a key requirement for the rational design of interventions aiming at enhancing EPC-mediated vascular repair (i.e. promoting reendothelialization and collateralization in the ischemic vascular bed) and reducing SMPC-mediated atherogenesis and development of restenosis in T2DM. The current research proposal is a next logical step to unravel these processes by performing numerical (flowcytometry and in vitro cell culture) and functional (in vitro cell culture and gene- and kinome profiling) analyses on vascular progenitor cells. To this end, vascular progenitor cells will be obtained from Type 2 diabetics with or without cardiovascular disease as well as from non-diabetics with or without cardiovascular disease.

Study objective

1) Determine differences in EPC and SMPC availability in the peripheral blood of T2DM patients with or without coronary artery disease (CAD) or peripheral artery disease (PAD) as well as in age- and sex-matched non-diabetic subjects with or without CAD or PAD.

2) Determine molecular and functional differences in EPCs and SMPCs between

these groups of patients.

These objectives will contribute to the identification of novel molecular targets for therapeutic intervention aiming at modulating vascular progenitor cell frequency and function in order to attenuate development of cardiovascular disease and restenosis in T2DM.

Study design

For this study six groups of patients will be included as summarized below:

Group 1: T2DM, + PAD, - CAD

Group 2: T2DM, - PAD, + CAD

Group 3: T2DM, - PAD, - CAD

Group 4: no T2DM, + PAD, - CAD

Group 5: no T2DM, - PAD, + CAD

Group 6: no T2DM, - PAD, - CAD

On the first next planned visit to the outpatient clinic of the Dept. Internal Medicine, Dept. Vascular Surgery or Cardiology of the UMCG, six extra tubes (total 60 ml) of peripheral blood will be obtained, in addition to the laboratory assessments (including HbA1c and lipid profile), which are routinely performed at each visit at the outpatient clinic. Peripheral blood samples will be collected in 6x 10ml EDTA vacutainer tubes (K3E 15% 0.12 ml). Since not all anticipated analyses can be performed simultaneously and because of limitations in obtainable cell numbers, patients will be requested to donate 60 ml of peripheral blood during two consecutive visits to the outpatient clinic. During the total duration of the study, each patient will donate 2x 60ml = 120 ml blood.

From a subgroup of patients included in group 1 (n=10) and 4 (n=10) (i.e. PAD patients with- or without T2DM, respectively) that will undergo endovascular surgery because of moderate to severe claudication (stage IIb-IV Fontaine classification) the atherosclerotic plaque will be collected. Prior to endarterectomy, 60 ml of peripheral blood will be collected. Immediately following endarterectomy the plaque will be divided into two parts: one part will be snap frozen, the other half will be formalin-fixed. Plaques will be used for histological analyses as well as gene/protein expression analyses.

Study burden and risks

The nature of the research is such that participation entails no inherent risk for the subject. The extent of the burden is relatively low (donation of 2x 60 ml peripheral blood during regular visits at the outpatient clinic of the UMCG) or donation of 60 ml blood prior to endovascular surgery. Healthy control subjects will come to our center specifically for the purpose of this study and

will therefore receive a modest compensation. Participants of the study will not have immediate benefit from the outcome of the study.

Contacts

Public

Universitair Medisch Centrum Groningen

Hanzeplein 1
PO Box 30.001 9700 RB Groningen
NL

Scientific

Universitair Medisch Centrum Groningen

Hanzeplein 1
PO Box 30.001 9700 RB Groningen
NL

Trial sites

Listed location countries

Netherlands

Eligibility criteria

Age

Adults (18-64 years)
Elderly (65 years and older)

Inclusion criteria

Patients eligible for participation are diagnosed with T2DM (duration 10-15 yrs) and divided into diabetic patients with PAD, with CAD or without cardiovascular disease. Patients eligible for participation in the various control groups are patients without a history of T2DM but with PAD or CAD, respectively. In addition, healthy individuals who have neither a history of diabetes, cardiovascular disease, inflammatory or autoimmune disease, nor obesity nor any other chronic disease are will be included.;Definitions PAD and CAD:;Peripheral artery disease (PAD)

PAD is defined by the presence of carotid stenosis or lower extremity atherosclerosis

obliterans. Severity of atherosclerosis of carotid vessels will be assessed by ultrasonography as a continuous variable. Diagnosis of atherosclerotic involvement of the lower extremities will be assessed by history of claudication or rest pain, bilateral pulses examination (dorsal pedal, posterior tibial, popliteal, and femoral arteries), ultrasonography performed bilaterally at levels of popliteal and femoral arteries, and, eventually, angiography. Patients are then classified according to the Leriche/Fontaine clinical classification of lower limb atherosclerosis obliterans.;Coronary artery disease (CAD)

CAD is defined as previous myocardial infarction, resting ECG indicative of past MI, positive ECG at exercise stress test, echocardiography stress-test positive for inducible ischemia, evidence of significant coronary artery stenose on angiography with or without typical chest pain. ;Participants allocated to the different groups will be matched for:

- gender
- age
- smoking
- BMI
- arterial hypertension
- HbA1c (diabetics)
- lipid profile

Exclusion criteria

Predefined exclusion criteria for T2DM include diagnosis of T1DM and presence of any of the following (self-reported) medical conditions:

- microvascular disease (diabetic retinopathy and nephropathy)
 - auto-immune diseases
 - current or prior cancers
 - acute or chonic infection
 - recent surgery or vascular intervention
 - age >80yrs
 - recent myocardial infarction
 - hemodialysis
 - immunosuppression
 - any other unrelated disease;
- Exclusion criteria for non-diabetics include:
- diagnosis of T1DM or T2DM
 - auto-immune diseases
 - current or prior cancers
 - acute or chonic infection
 - recent surgery or vascular intervention
 - age >80yrs
 - recent myocardial infarction
 - hemodialysis
 - immunosuppression
 - any other unrelated disease

Study design

Design

Study type:	Observational non invasive
Intervention model:	Other
Allocation:	Non-randomized controlled trial
Masking:	Open (masking not used)
Control:	Active
Primary purpose:	Basic science

Recruitment

NL	
Recruitment status:	Recruitment stopped
Start date (anticipated):	16-01-2010
Enrollment:	180
Type:	Actual

Ethics review

Approved WMO	
Date:	13-02-2009
Application type:	First submission
Review commission:	METC Universitair Medisch Centrum Groningen (Groningen)
Approved WMO	
Date:	11-05-2010
Application type:	Amendment
Review commission:	METC Universitair Medisch Centrum Groningen (Groningen)

Study registrations

Followed up by the following (possibly more current) registration

No registrations found.

Other (possibly less up-to-date) registrations in this register

No registrations found.

In other registers

Register	ID
CCMO	NL25792.042.08