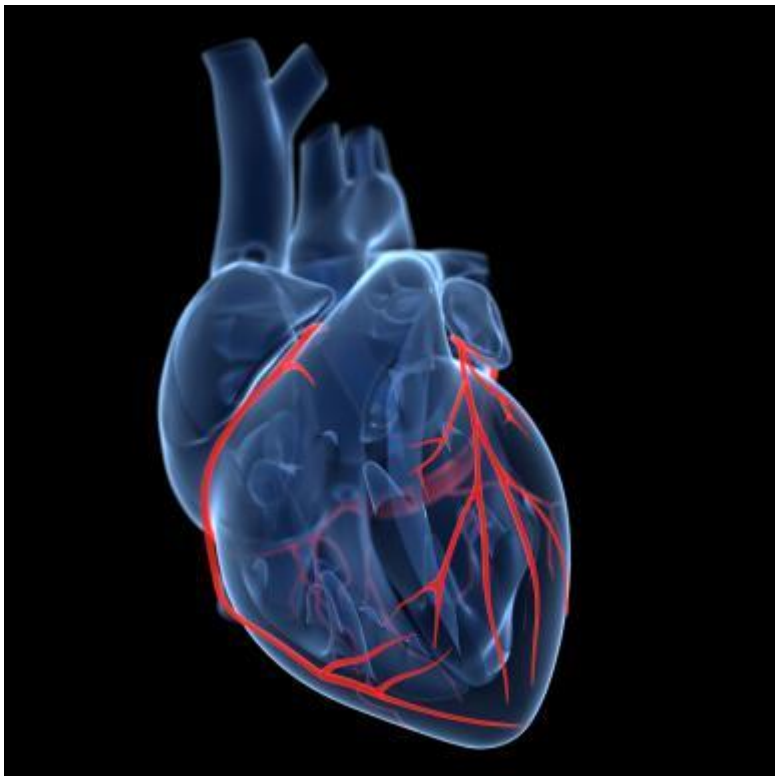
Student thesis

Abnormal glucose regulation and coronary artery disease



Medical student: Siril Haug

Supervisors: Erik Madssen and Rune Wiseth

NTNU, Det Medisinske Fakultet
Norwegian University of Science and Technology,
Faculty of Medicine

Table of contents

Summary of thesis

Abnormal glucose regulation describes a condition with disturbed glucometabolic status. Patients with this abnormality have an increased risk of developing both micro- and macrovascular complications. Patients with abnormal glucose regulation composite a large percentage of patients undergoing coronary revascularization. These patients have a poorer prognosis following coronary revascularization compared to normoglycaemic patients. Thus, screening for undiagnosed diabetes mellitus is recommended in patients with established coronary artery disease. Currently there are three methods available to identify abnormal glucose regulation; fasting plasma glucose, an oral glucose tolerance test and glycated haemoglobin (HbA1c). The sensitivity of these methods in identifying abnormal glucose regulation in patients with concomitant coronary artery disease is essential. We therefore aimed to validate the diagnostic strength of in-hospital HbA1c in detection of abnormal glucose tolerance in patients undergoing PCI with an oral glucose tolerance test 4-6 weeks after index PCI as gold standard. Our study included eighty-six patients from the Gluko-Norstent study, which is a trial designed to analyse the association between glucometabolic disturbances and the long-term outcome after PCI with stent implantation. A comprehensive review of the literature was performed as part of this project. When using the recommended cut off value of HbA1c of 6.5 % we found that abnormal glucose regulation was underdiagnosed with in-hospital HbA1c when compared to an oral glucose tolerance test. The "high risk" HbA1c range of 5.7-6.4 % seemed to overestimate the number of patients with impaired glucose tolerance based on the oral glucose tolerance test. The combination of in-hospital fasting plasma glucose and HbA1c was found to add diagnostic strength in identifying abnormal glucose tolerance in patients with established coronary artery disease.

1. Introduction

1.1 Abnormal glucose regulation

Abnormal glucose regulation refers to impaired glucose tolerance (IGT), impaired fasting glucose (IFG), "high-risk" HbA1c and diabetes mellitus (DM). DM describes a group of metabolic diseases characterized by hyperglycaemia, resulting from relative or absolute defects in insulin secretion, insulin action, or both [1]. IGT and IFG, also referred to as prediabetes or intermediate hyperglycaemia, are not clinical entities, but rather risk factors for development of DM [2, 3]. Similarly, HbA1c values below the diagnostic threshold for DM, referred to as "high-risk" HbA1c, has been proposed to identify patients at high risk of developing DM [2, 4].

Persistent hyperglycaemia causes reduced function in several organs, resulting in diabetes-specific complications like neuropathy, nephropathy and retinopathy [5]. These patients also have an increased risk of developing macrovascular disease, such as coronary artery disease (CAD), peripheral artery disease and cerebrovascular disease [6].

The World Health Organization (WHO) and the American Diabetes Association (ADA) provide recommendations on classification and diagnostic criteria of DM and other groups of abnormal glucose regulation. The latest recommendations from WHO are summarized in Table 1. The level of glycaemia associated with the development of diabetes-specific retinopathy, has been agreed upon as suitable diagnostic cut off values for DM [7, 8]. This is based on large epidemiological data showing that the prevalence of diabetes-specific retinopathy increases in a linear fashion with increasing glycaemic values [9]. Regarding prediabetes, there is some controversy between WHO and ADA as to which criteria that defines this group [2, 10].

Table 1: Recommendations on diagnosis of diabetes mellitus and other disturbances in glucose regulation (WHO 2011):	
Diabetes mellitus* (Either 1, 2, 3 or 4)	1. HbA1c ≥ 6.5 % 2. FPG ≥ 7.0 mmol/l 3. 2h PG ≥ 11.1 mmol/l 4. Symptoms of hyperglycaemia and random plasma glucose ≥ 11.1 mmol/l
Intermediate hyperglycaemia/prediabetes:	
Impaired glucose tolerance	FPG <7.0 mmol/l and 2hPG ≥ 7.8 and <11.1 mmol/l
Impaired fasting glucose	FPG ≥ 6.1 and <7.0 mmol/l and (if measured) 2hPG <7.8 mmol/l

*In the absence of symptoms, test method 1, 2 or 3 has to be positive two subsequent times to give the diagnosis of DM.

In 2014 the prevalence of DM was estimated to 387 million [11], with a substantial number of people with type 2 DM still unaware of their disease. The global burden of DM continues to grow, with a worldwide prevalence expected to reach 592 million by 2035 [12]. The increasing prevalence emphasizes the importance of detection and treatment of people with DM and people with high risk of future DM.

1.2 Coronary artery disease and abnormal glucose regulation

Atherosclerosis is the principal cause of CAD. Currently, atherosclerosis is considered a chronic inflammatory continuum that selectively affects arteries, occurring far more often in patients with cardiovascular risk factors such as a high concentration of lipoproteins rich on cholesterol, cigarette smoking, DM, and hypertension [13]. With aging and in the presence of these factors, the endothelium lining the arterial wall gets dysfunctional, promoting the atherosclerotic process. The development of atherosclerosis is summarized in Figure 1, modified from Weber et al. [14].

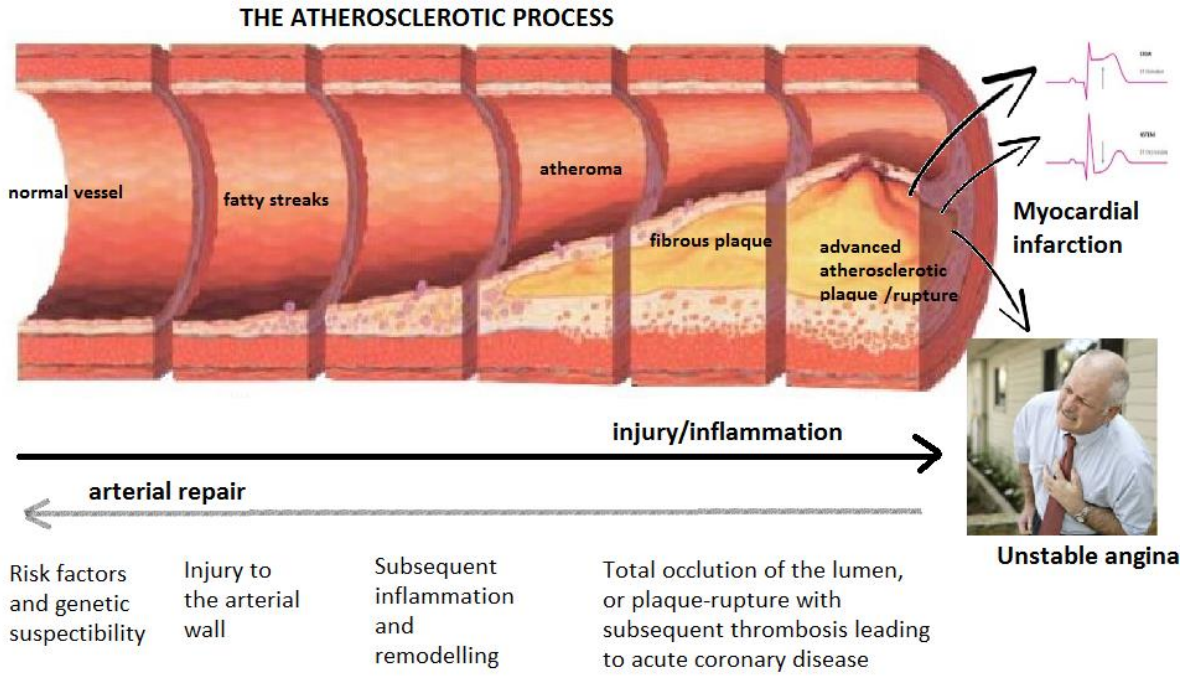


Figure 1): *The process of atherosclerosis. In the presence of risk factors and genetic susceptibility factors, an injury to the endothelium leads to endothelial dysfunction and loss of endothelial integrity. This leads the way for noxious cells and molecules into the subendothelial layers of the arterial wall. Inflammatory cells colonize the arterial wall, and initiate a proinflammatory state. Remodelling and proliferation of cells form an atheroma, which reduces the vessel lumen. The atherosclerotic plaque might progressively occlude lumen, or rupture with subsequent thrombosis resulting in acute coronary disease (myocardial infarction or unstable angina).*

Patients with abnormal glucose regulation are prone to an accelerated atherosclerotic process, characterized by a pro-thrombotic and a pro-inflammatory state [15-17]. Factors associated with abnormal glucose regulation (Figure 2) will alter the function of cells and the function of the vessel wall, thus resulting in promoted atherosclerosis.

Dyslipidaemia associated with abnormal glucose regulation often involves smaller and denser LDL molecules, which are more susceptible to oxidation and therefore promote development of atherosclerosis. Hyperglycaemia increases the production of reactive oxygen species, contributing to the atherosclerotic process [16]. Furthermore, chronic hyperglycaemia results in glycation of several macromolecules, producing Advanced Glycation End Products (AGEP). Oxidation of AGEPs seems to be important in initiating the oxidation of lipids, contributing to the increased atherosclerotic process in DM [18].

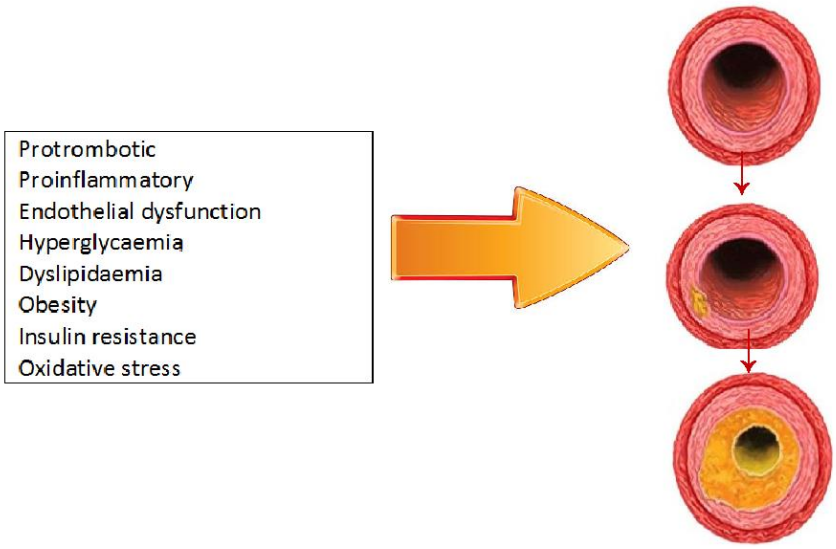


Figure 2) *Key features in patients with abnormal glucose regulation, which leads to an accelerated atherosclerotic process.*

Studies using coronary angiography have demonstrated that patients with DM have a more extensive and diffuse CAD compared to patients without DM, and that accelerated coronary atherosclerosis is present already at a prediabetic stage [19, 20].

Coronary atheromas are often referred to as stable or unstable plaques, based on the plaque composition (Figure 3). Atheromas may cause clinical symptoms or events by progressive narrowing of the vessel lumen, or by rupture or erosion of the plaque (Figure 1). Unstable plaques have an increased risk of rupture or erosion and for subsequent thrombosis, causing acute coronary syndromes [21].

Coronary plaques in patients with abnormal glucose regulation has shown to contain more lipid and less fibrous tissue, compared to subjects with normal glucose regulation [20]. These features are considered the hallmark of vulnerable plaques with an increased risk of plaque rupture or erosion and subsequently an acute coronary syndrome.

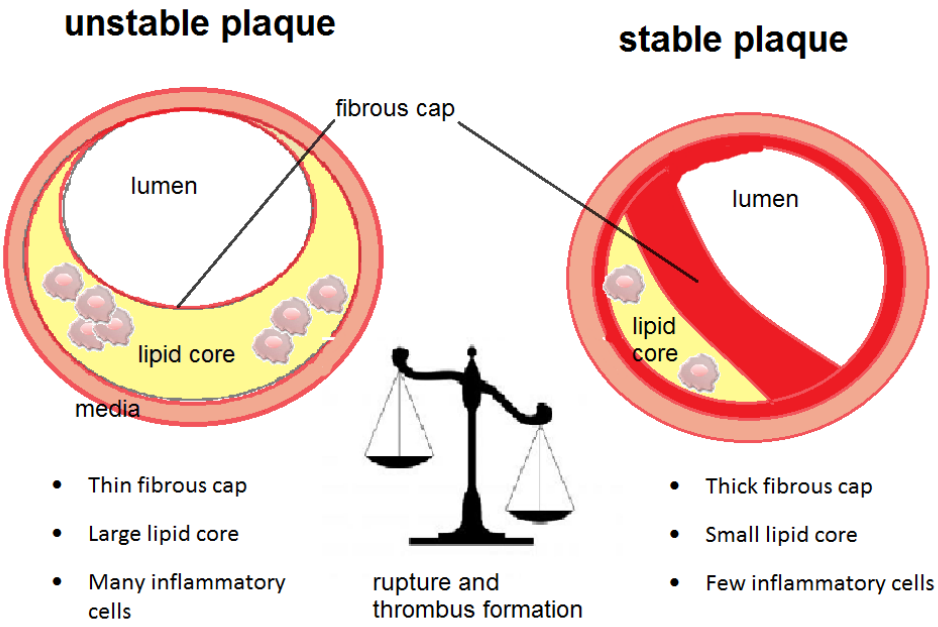


Figure 3) *Morphological differences in stable and unstable atherosclerotic plaque. Unstable plaques are more susceptible for plaque rupture or erosion, and subsequent thrombosis.*

1.3 Coronary revascularization in patients with abnormal glucose regulation

Randomized studies of patients with DM and stable CAD have shown no advantages of coronary revascularization combined with optimal medical treatment, compared to optimal medical treatment alone, regarding the mortality rates and the risk of non-fatal myocardial infarction [22, 23]. The European Society of Cardiology recommends that optimal medical treatment should be considered as the preferred treatment in patients with DM and stable CAD [24].

Nevertheless, a significant proportion of patients with DM and concomitant stable CAD undergo coronary revascularization due to symptoms of ischemia (i.e. angina pectoris) or silent ischemia established by functional testing. Patients with DM are prone to silent ischemia due to diabetes associated autonomic neuropathy, which is related to adverse cardiac outcome [25].

1.3.1 PCI versus CABG surgery

Approximately one fourth of coronary revascularization procedures are performed in patients with DM. Despite advantages in the treatment of CAD, patients with abnormal glucose regulation have a less favourable outcome after coronary revascularization. A higher rate of repeated revascularization procedures after PCI with stent implantation has been consistently found in patients with DM compared to patients without DM, included in randomized clinical trials comparing PCI with stent implantation and CABG surgery [26-29]. These results correspond to recommendations from the European Society of Cardiology stating that CABG surgery is superior to PCI with stent implantation in patients with DM and stable multi-vessel CAD, but only in patients with an acceptable surgical risk [30].

1.3.2 Restenosis and stent thrombosis

Although CABG is the preferred treatment for patients with DM and stable multi-vessel CAD, a significant proportion of patients with DM undergo PCI with stent implantation due to co-morbidities, the presence of single-vessel CAD or if presented as an acute coronary syndrome. Following stent implantation, patients with abnormal glucose regulation are at a higher risk of restenosis and stent-thrombosis compared to patients without DM [31, 32].

Restenosis is a gradual narrowing of the stented segment, caused by arterial damage from the stent implantation and subsequent neointimal tissue proliferation [33]. In comparison, stent-thrombosis is a sudden thrombotic occlusion of the stent, most often resulting in a clinical presentation with an acute myocardial infarction or sudden death. Stent thrombosis is most common within the first month after stent implantation, but may also occur more than one year after the procedure.

Patients with DM usually have a more diffuse CAD, smaller vessel diameter, a greater length of the stented segment, an increased atherosclerotic process and an exaggerated neointimal hyperplasia, all of which increase the risk of restenosis following stent implantation [34]. Additionally, hyperglycaemia increases platelet activation and adhesion, increasing the risk of stent-thrombosis.

The development of first-generation drug-eluting stents significantly reduced the rate of restenosis compared to bare-metal stents in patients with DM, consistent with the effect on the overall population [35-39]. Drug-eluting stents releases an anti-proliferative drug, thus reducing the rate of restenosis.

Second-generation drug-eluting stents are now the preferred choice of stent implantation for patients with DM [40]. Newer generations of stent may further reduce the risk of restenosis and stent thrombosis. For instance, bioresorbable polymer drug-eluting stents may be as safe and efficacious as second-generation drug eluting stents [41].

1.4 Diagnostic methods in detection of abnormal glucose tolerance

Traditionally, DM has been diagnosed by measuring venous plasma glucose levels; either FPG, randomly measured plasma-glucose or 2-hour plasma glucose (2hPG) after ingestion of 75 g oral glucose load (OGTT). Recently, HbA1c was introduced as a diagnostic test for DM [2, 3]. All methods are currently in use, although there is some controversy as to which method is preferred [42].

1.4.1 Venous plasma glucose measurements

Measurement of venous plasma glucose in fasting subjects and after an oral glucose load of 75 grams is widely accepted as a diagnostic criterion for diabetes [1, 2]. The OGTT evaluates the efficiency to metabolize glucose in the body. Increased postprandial glucose concentration usually occur before the FPG increases, and the OGTT is therefore a sensitive method for detecting patients at risk of developing DM and an early marker of abnormal glucose regulation [43]. However, the OGTT requires extensive patient preparation and is often impractical due to the method being time-consuming.

Numerous factors might influence the venous plasma glucose concentration, resulting in a high intraindividual variability and low reproducibility of FPG-measurements and oral glucose tolerance testing [43].

1.4.2 HbA1c

HbA1c refers to the glycated haemoglobin, i.e. the percentage proportion of haemoglobin-molecules attached to a glucose molecule [44]. The glucose molecule will stay attached to haemoglobin until the erythrocytes are renewed. The average survival-time for erythrocytes in blood is 120 days, or 8-12 weeks. Consequently, HbA1c levels can be used to reflect the

average plasma glucose levels during the last 8-12 weeks [45], and is therefore useful in detection of long-term blood-glucose levels and disease monitoring of patients with DM.

Evidence for the use of HbA1c in monitoring glycaemic control in patients with DM is based manly in its association with development of diabetes-specific retinopathy in observational studies [9, 46].

In 2009, an International Expert Committee recommended that HbA1c should be used as a diagnostic tool in DM with a diagnostic cut off value of ≥ 6.5 % [4]. This was based on a strong correlation between HbA1c and retinopathy, an acceptable accuracy and precision of the HbA1c assay, and also a low biological- and preanalytical variability compared to plasma glucose measurements. ADA and WHO added the HbA1c ≥ 6.5 % as a diabetes-criterion in 2010 and 2011, respectively [2, 10].

1.4.3 HbA1c versus venous plasma glucose measurements

From a clinical perspective, HbA1c has several advantages compared to a standard glucose measurement. The blood sample can be taken at any time of the day, it requires no special patient preparation (i.e. fasting) and it is less time consuming than performing an oral glucose tolerance test. Compared to the plasma glucose measurement, HbA1c has a lower biological- and preanalytical variability [43], and it is unaffected by day-to-day variations in plasma glucose concentrations.

Although the HbA1c assay shows numerous favourable abilities compared to standard glucose measurements, there are some limitations concerning the diagnostic use of HbA1c (Table 2, adapted from Gallagher et at. [47]). In these situations, clinicians should use venous glucose measurements.

HbA1c levels are known to increase with normal ageing [48], and studies have shown ethnic differences in HbA1c levels [49]. The reason and significance of these ethnic- and age-concerning differences are not established, accordingly there are no current ethnic- or age-specific diagnostic HbA1c values.

Table 2. Factors that might influence the HbA1c measurement

Erythropoiesis	Increased HbA1c can be seen in iron- and vitamin B12-deficiency and in decreased erythropoiesis. Decreased HbA1c levels can occur in erythropoietin-, iron- and Vitamin B12-treatment, and in chronic liver disease.
Altered haemoglobin	Fetal haemoglobin, different hemoglobinopathies and genetic determinants may give variable change in HbA1c levels.
Glycation	Increased HbA1c levels in alcoholism, chronic renal failure etc. Decreased HbA1c levels with ingestion of vitamin C and E, or aspirin, and more.
Erythrocyte lifespan	All conditions with prolonged erythrocyte lifespan will give increased HbA1c, for example splenectomy. In contrast, decreased HbA1c is seen with shortened erythrocyte lifespan, like in hemoglobinopathies, splenomegaly, different drugs, and rheumatic arthritis.
HbA1c assays	Different conditions may interfere with different HbA1c assays, like hyperbilirubinemia, hypertriglyceridemia, and some hemoglobinopathies.

Several large population-based studies have shown that the diagnostic cut off value of HbA1c ≥ 6.5 % results in a high specificity, but a low sensitivity for detecting DM compared to the traditional glucose measurements [50-58]. Thus, a high proportion of individuals with DM based on venous plasma glucose measurements, might be incorrectly classified as non-diabetic based on the HbA1c assay alone. Conversely, it is conceivable that the HbA1c assay might identify a larger proportion of patients with DM than venous plasma glucose measurement, as the test is more convenient in the clinical setting.

Furthermore, studies have demonstrated that different people are diagnosed with DM, when the HbA1c assay or the venous plasma glucose measurements are used independently [50-52, 57]. Table 3 summarizes the benefits and disadvantages of HbA1c compared to venous glucose measurements in diagnosing abnormal glucose regulation.

Table 3. Advantages and disadvantages of HbA1c, FPG and the OGTT

HbA1c	**FPG**	**OGTT**
Advantages: - No patient preparation - Can be taken any time of the day - Low biological variability - Not altered by stress and acute illness - Reflects long-term blood glucose levels - Standardized method - Correlates with development of retinopathy - Highly specific for detection of DM Disadvantages: - May be influenced by numerous factors (Table 2) - Not universally available - Low sensitivity for detection of DM - Low sensitivity in detection of patients with high risk of future DM	Advantages: - Available - Inexpensive - Easily performed - Higher reproducibility than the OGTT Disadvantages: - Requires fasting - Large biological variability - Diurnal variation - Influenced by stress and acute illness - Low sensitivity in detecting DM compared to OGTT - Low sensitivity in detection of patients at high risk of developing DM (i.e. IGT)	Advantages: - Highly sensitive in detection of DM - Sensitive indicator on risk of developing DM (i.e. IGT) - Standardized assay Disadvantages: - Low reproducibility - Extensive patients preparation - Time-consuming - Preanalytical and analytical variability is high

1.5 Objectives

The NorStent study is a large randomized trial in patients with CAD treated with stent implantation, designed to compare the long-term effect of drug-eluting stents versus bare-metal stents [59]. The Gluko-Norstent study includes a subgroup of participants form the NorStent study. This subgroup underwent HbA1c- and FPG-measurements during admission for PCI-treatment, with a purpose to analyse the association between glucometabolic disturbances (based on HbA1c measurements during admission) and the long-term outcome after stent implantation. However, in order to obtain a classification of glucometabolic disturbances being as correct as possible, an OGTT is required. Therefore, eighty-six patients from the Gluko-NorStent trial performed an OGTT 4-6 weeks after the index-PCI. Our aim was to validate the diagnostic strength of in-hospital HbA1c in detection of abnormal glucose regulation compared to an OGTT at follow-up. The reproducibility of fasting plasma glucose measured during hospitalisation and at follow-up was also assessed.

2 Materials and methods

2.1 Study participants

The present study included 86 patients included in the NorStent trial. The patients had no previous diagnosis of DM. All study participants underwent coronary angiography and PCI between October 2009 and March 2011 at St.Olav Hospital in Trondheim. All patients gave informed consent. The study protocol was approved by the Regional Committee for Medical Research Ethics (REK) in Middle-Norway.

2.2 Laboratory tests

Arterial plasma glucose was measured during the PCI-procedure, and HbA1c was measured during hospitalization. Venous FPG was measured after overnight fasting the day following PCI. Trained nurses measured venous FPG and performed the OGTT 4-6 weeks after the PCI-procedure. A standardized OGTT was performed according to the recommendation of the WHO [60], by measuring the venous plasma glucose level two hours after an oral glucose load of 75 grams of glucose. Plasma levels of HbA1c were analysed by an immunological method using Roche Cobas Integra 400 (F. Hoffmann-La Roche Ltd, Switzerland). HbA1c levels are reported as percentages (%). All plasma glucose concentrations were analysed with Roche Modular P. (F. Hoffmann-La Roche Ltd, Switzerland).

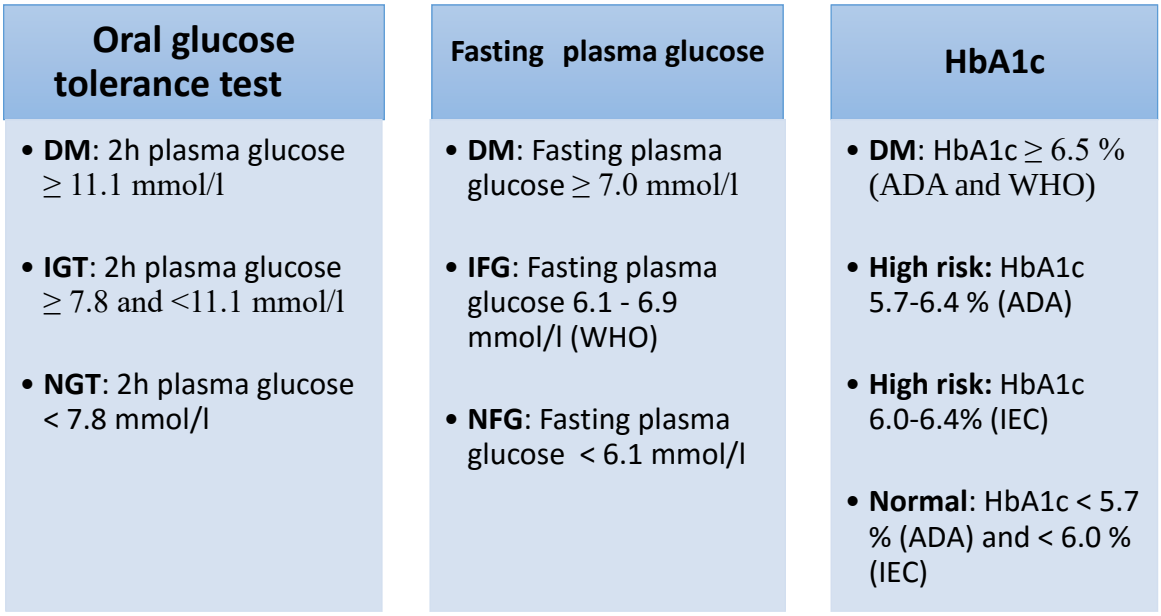
2.3 Classification of abnormal glucose regulation

Patients were categorized in groups based on different methods of detecting abnormal glucose regulation (Figure 4). A cut off value of HbA1c $\geq$ 6.5 % for diagnosing DM was used [10]. Patients at "high risk" of developing DM were identified by HbA1c in the ranges of 5.7-6.4 % and 6.0-6.4 %, according to statements from the American Diabetes Association(ADA) and an International Expert Committee, respectively [2, 4]. Based on the standardized OGTT, patients were categorized into three groups; DM, IGT and normal glucose tolerance (NGT) [1]. Based on FPG the patients were categorized into normal fasting glucose (NFG), impaired fasting glucose (IFG) and DM.

In the analyses, patients were divided into three groups: (1) abnormal glucose tolerance including patients with DM and IGT based on OGTT, (2) abnormal fasting glucose including

patients with DM and IFG based on FPG, and (3) abnormal HbA1c including patients with high-risk HbA1c and DM based on HbA1c.

Figure 4. Categorization of abnormal glucose regulation



ADA; American Diabetes Association. WHO; World Health Organization. IEC; International Expert Committee.

2.4 Statistical analysis

Categorical data are presented as proportions, and continuous data as medians with 25 and 75 percentiles in parenthesis due to non-normal distribution of many variables. Categorical variables were compared with the Chi-square test. For continuous data, within-group comparisons were performed with Wilcoxon's signed rank test and between-group comparisons were performed with the Mann-Whitney *U* test. P-values <0.05 were considered statistically significant. The package IBM SPSS Statistics for Windows (version 21.0, Armonk, NY, IMB Corp.) was used for all statistical analysis.

3 Results

Eighty-six patients, 66 men and 20 women, in the age between 39-79 years, were included. The median HbA1c level was 5.8 % (5.6, 6.0), the median FPG during hospitalisation was 5.5 mmol/l (5.0, 6.2), the median FPG at 4-6 weeks was 5.8 mmol/l (5.3, 6.1) and the median 2h plasma glucose after an OGTT was 6.4 mmol/l (5.1, 8.4). Table 4 presents patients' characteristics stratified by normal glucose tolerance and abnormal glucose tolerance defined by the OGTT. Patients with abnormal glucose tolerance based on the OGTT was significantly older and had significantly higher levels of HbA1c, in-hospital FPG and FPG at follow up.

Table 4: Characteristics of patients according to glucometabolic category defined by OGTT at 4-6 weeks

	NGT (n=60)	AGT (n=26)	p-value
Age (years)	61 (56, 67)	65 (61, 73)	0.037
HbA1c (%)	5.8 (5.6, 5.9)	5.9 (5.7, 6.3)	0.007
Plasma glucose during PCI-procedure (mmol/l)	5.5 (5.0, 6.3)	6.0 (5.0, 7.2)	0.157
FPG in-hospital (mmol/l)	5.4 (5.0, 5.8)	6.2 (5.6, 6.7)	<0.001
FPG at 4-6 weeks (mmol/l)	5.6 (5.2, 5.9)	6.2 (5.8, 6.7)	<0.001

3.1 Identification of abnormal glucose regulation

3.1.1 HbA1c versus OGTT

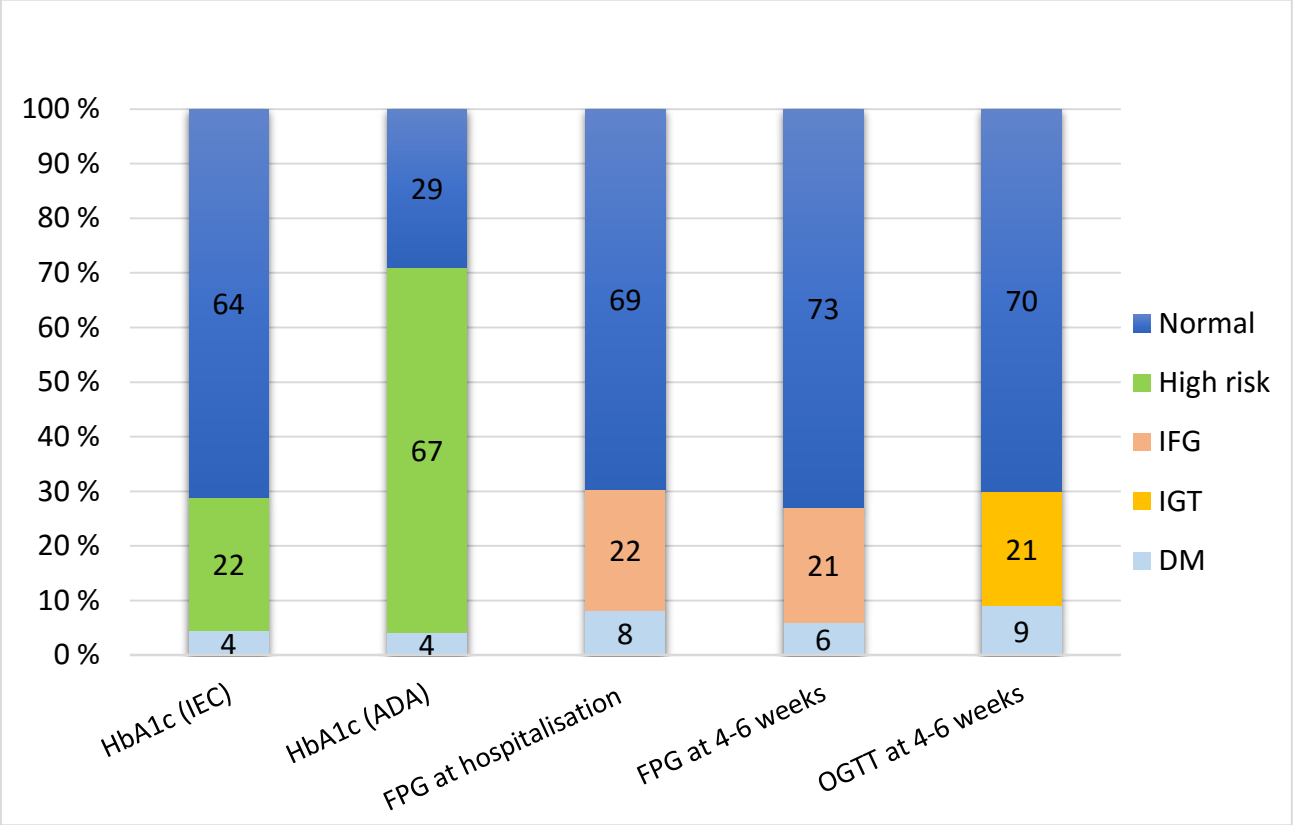
Based on HbA1c results, DM was diagnosed in 4 %, while 67 % and 22 % were identified as "high risk" using the proposed HbA1c-ranges of 5.7-6.4 % and 6.0-6.4%, respectively. Based on the OGTT at follow-up, DM was diagnosed in 9 % of patients; IGT in 21 %, and NGT in 70 % (Figure 5). 71 % had an abnormal HbA1c using the cut off value ≥ 5.7 %, while 26 %

had an abnormal HbA1c when using the cut of value ≥ 6.0 % (Figure 6). In comparison, 30 % was diagnosed with abnormal glucose tolerance based on the OGTT.

34 % with an HbA1c ≥ 5.7 % were identified as having abnormal glucose tolerance as detected by the OGTT (Figure 7). There was no significant association between an HbA1c level ≥ 5.7 % and the results following the OGTT performed at follow-up, in terms of detecting abnormal glucose tolerance (χ^2 (1) = 1.75, p= 0.19). In comparison, 50 % of patients with an HbA1c ≥ 6.0 % had abnormal glucose tolerance based on the OGTT. The association between an HbA1c level ≥ 6.0 % and abnormal glucose tolerance based on the OGTT was significant (χ^2 (1) = 5.5, p= 0.019).

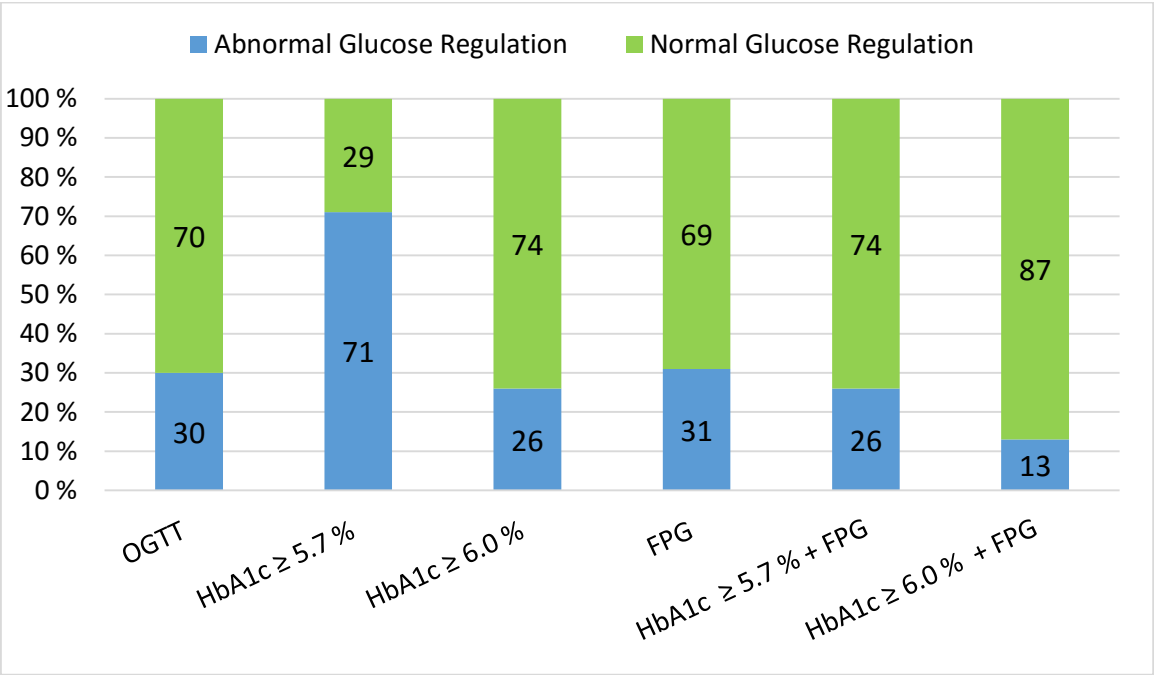
Figure 8 illustrates the distribution (%) of glucometabolic groups based on the OGTT among patients with high-risk HbA1c levels. Among patients identified as “high risk” with an HbA1c in the range 5.7-6.4 %, 12 patients (21 %) were identified as having IGT based on the OGTT performed at follow-up. 40 patients (69 %) were identified as having NGT, while 6 patients (10 %) were diagnosed with DM based on the OGTT. In comparison, of patients with an HbA1c in the range of 6.0-6.4 %, 4 patients (21 %) were diagnosed with IGT, 11 patients (58 %) with NGT and 4 patients (21 %) with DM.

Figure 5 Classification of patients based on different tests



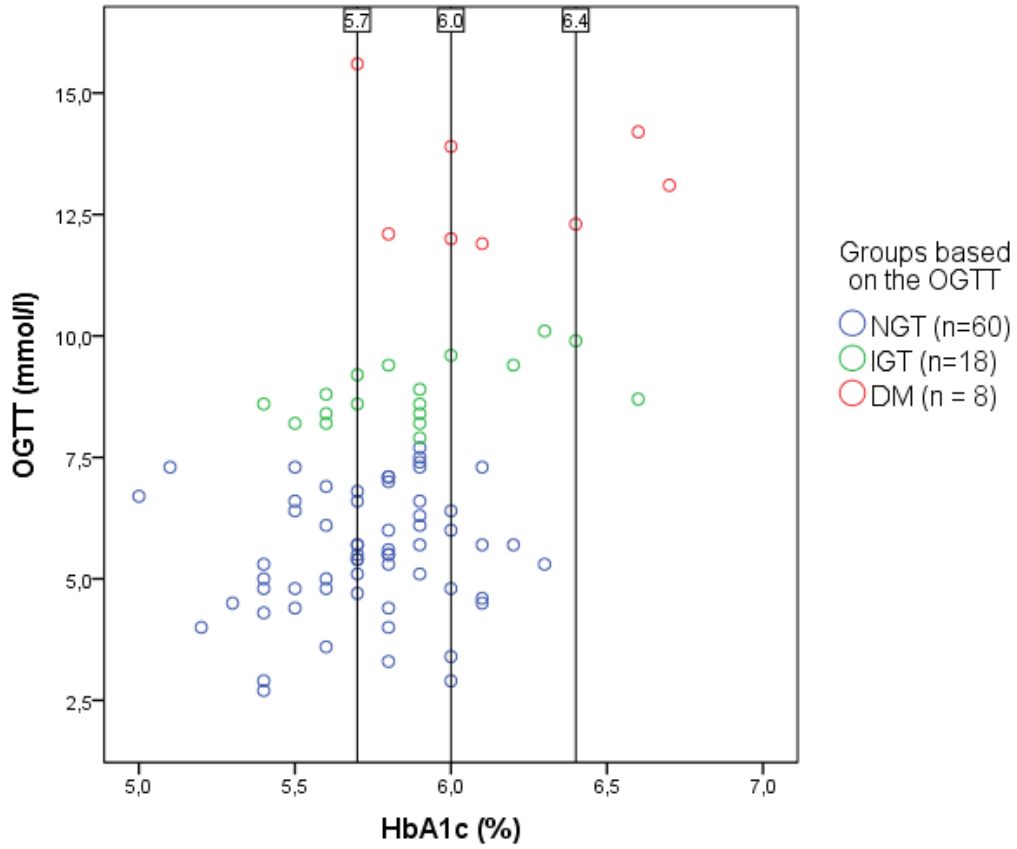
High risk; HbA1c levels in the range of 6.0-6.4% (IEC) and 5.7-6.4 % (ADA).

Figure 6 Abnormal glucose regulation versus normal glucose regulation classified by different test



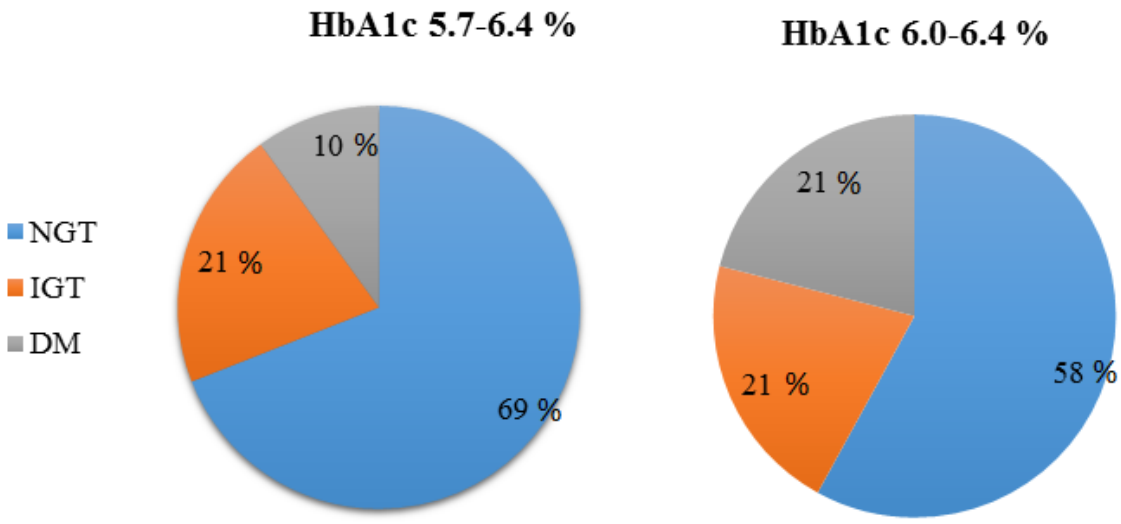
FPG; FPG measured during hospitalisation for PCI treatment.

Figure 7 Scatterplot illustrating the relationship between HbA1c and venous glucose concentration after an OGTT



The vertical lines illustrate the proposed range of HbA1c values; high risk indicates HbA1c in the range 5.7-6.4 % and 6.0-6.4%.

Figure 8 Distribution (%) of glucometabolic groups based on the OGTT among patients with "high risk" HbA1c levels



3.1.2 HbA1c versus FPG

In-hospital FPG identified 26 patients (31 %) with abnormal fasting glucose. 22 patients (37 %) with HbA1c ≥ 5.7 % had an abnormal fasting glucose based on the FPG. In comparison, 11 patients (50 %) with abnormal HbA1c ≥ 6.0 % had abnormal fasting glucose based on the FPG test. The association between HbA1c ≥ 6.0% and abnormal fasting glucose was significant (χ^2 (1) = 5.3, p= 0.022).

3.1.3 FPG vs OGTT

In-hospital FPG identified 7 patients with DM, 19 with IFG and 59 with NFG (Figure 5), compared to 8 patients with DM, 18 with IGT and 60 with NGT based on the OGTT. 16 patients (62 %) with abnormal fasting glucose had abnormal glucose tolerance based on the OGTT. The association between abnormal fasting glucose detected by in-hospital FPG and abnormal glucose tolerance based on the OGTT at follow-up was significant (χ^2 (1) = 16.9, p < 0.001).

3.1.4 HbA1c and FPG versus OGTT

22 patients (26 %) had the combination of abnormal HbA1c level ≥ 5.7 % and abnormal fasting glucose. 14 patients (64 %) with HbA1c ≥ 5.7 % and abnormal fasting glucose had abnormal glucose tolerance based on the OGTT. Correspondingly, 11 patients (13 %) were identified with abnormal HbA1c ≥ 6.0 % and abnormal fasting glucose. 9 patient (82 %) with HbA1c ≥ 6.0 % and abnormal fasting glucose had abnormal glucose tolerance based on the OGTT. The combination of abnormal HbA1c and abnormal FPG had a significant association with abnormal glucose tolerance based on OGTT (χ^2 (1) = 15.6, p <0.001 and χ^2 (1) = 15.9, p <0.001, by using HbA1c cut off value ≥ 5.7 % and ≥ 6.0 % respectively).

3.1.5 Reproducibility of FPG

In-hospital FPG classified 7 patients (8 %) as having DM, 10 patients (22 %) with IFG and 59 patients (69 %) with NFG. One patient did not perform an FPG during hospitalisation. Correspondingly, FPG at follow-up identified 5 patients (6 %) with DM, 18 patients (21 %) with IFG and 63 patients (73 %) with NFG (Figure 5). 59 patients (69 %) remained in the same FPG-category after a repeat FPG measurement. 47 patients (55 %) measured a higher FPG at follow-up than FPG measured during hospitalisation, while 29 patients (34 %) measured a lower FPG value at follow-up. The difference between in-hospital FPG and FPG at follow-up was not significant (Z= - 1.840, p=0.066).

4 Discussion

In the present study, we found that in-hospital HbA1c ≥ 6.5 % underdiagnosed patients with DM as detected by an OGTT after 4-6 weeks. Furthermore, we found a poor association between HbA1c values ≥ 5.7 % and glucose tolerance status defined by the OGTT. HbA1c values in the range 5.7-6.4 % seem to overestimate the risk of having IGT, while HbA1c 6.0-6.4 % performed better at identifying patients with IGT detected by the OGTT. The combination of HbA1c and FPG added diagnostic strength compared to HbA1c used solely, in terms of identifying abnormal glucose regulation confirmed by an OGTT. Furthermore, FPG during hospitalisation seem to reproduce sufficiently after 4-6 weeks.

We found a lower prevalence of newly diagnosed DM by using the HbA1c criteria ≥ 6.5 % compared to the OGTT (3.5 % versus 9.3 %). HbA1c failed to detect 6 patients (75 %) with DM according to the results from the OGTT. These results are consistent with other studies in patients with CAD [61-63], and with results from a large study comparing FPG, OGTT and HbA1c in patients with CAD [64]. Large populations-based studies have shown that HbA1c has a high specificity, but low sensitivity in diagnosing DM compared to an OGTT [54, 56], resulting in a high number of patients that are incorrectly classified as non-diabetic based on HbA1c solely.

Conversely, it has been argued that the HbA1c assay might identify a larger proportion of patients with DM than the OGTT, due to the convenience of the HbA1c test in the clinical setting. This might be true when screening for abnormal glucose regulation in a general population. However, underdiagnosing abnormal glucose regulation in patients with CAD may have essential impact on the prognosis and efforts should therefore be made to minimize the risk of missing the diagnosis of dysglycemia [65]. A sensitive method for detecting abnormal glucose regulation, like the OGTT, is therefore of great importance regarding assessment of cardiovascular risk in patients with established CAD. Furthermore, studies comparing all three methods for detecting abnormal glucose regulation have shown that 2-hour plasma glucose has the strongest association with mortality and risk of cardiovascular disease [66, 67].

The recommended diagnostic cut off value of HbA1c ≥ 6.5 % for DM, initially proposed by an expert committee, was based on the association between HbA1c levels and the occurrence of diabetes-specific retinopathy [4]. Due to the continuous risk for development of DM, occurring within a wide range of HbA1c levels, the expert committee did not formally identify a subdiabetic HbA1c-range that classifies patients in categories similar to IGT and IFG. However, they noted that subjects with HbA1c within the range 6.0-6.4 % are at very high risk of developing DM. Inconsistently, the ADA proposed that HbA1c levels in the range 5.7-6.4 % performs better at identifying high-risk individuals, arguing that HbA1c values in the range 6.0-6.4 % fails to identify a substantial number of patients with IGT and/or IFG [2].

Our results indicate that HbA1c values in the range 5.7-6.4 % seem to overestimate the proportion of patients with IGT based on the OGTT, yielding a high percentage of false positive results. We found that 58 patients (67 %) had an HbA1c in the range 5.7-6.4 %, of which 12 patients had IGT based on an OGTT (Figure 8). This is consistent with results in other studies [62, 68], showing that HbA1c has a high specificity but low sensitivity in detecting IGT compared to the OGTT. Furthermore, we found that a high-risk HbA1c range of 6.0-6.4 % resulted in a distribution of categories more similar to those based on the OGTT criteria.

In our study, in-hospital FPG identified 7 patients with DM, compared to 8 patients diagnosed with DM based on the OGTT. Also, we found that classification of patients in different glucometabolic groups based on in-hospital FPG and OGTT at follow-up corresponded well (Figure 5 and 6).

However, several studies have shown that FPG ≥ 7.0 mmol/l lack sufficient sensitivity in screening for DM compared to an OGTT [69, 70], and FPG used independently would incorrectly classify patients as non-diabetic. A large study in patients with CAD found that evaluation of glucometabolic status based solely on FPG misclassified a substantial number of patients with DM and IGT as detected by an OGTT [71]. To identify patients with IGT is of clinical significance, as patients with IGT are more prone to progression of CAD compared to patients with IFG [72].

Neither FPG nor HbA1c has proven sufficient sensitivity for detection of patients with DM, compared to the OGTT. However, considering the extensive patient preparation and the

considerable time consumption, an OGTT is often not feasible. Furthermore, performing an OGTT in patients with acute coronary syndrome will most often be impractical. Therefore, the combination of HbA1c and FPG has been proposed as an alternative to the OGTT [40].

We found that the combination of HbA1c ≥ 5.7 % and FPG ≥ 7.0 mmol/l was better at identifying patients with abnormal glucose tolerance based on an OGTT, compared to HbA1c alone. This is in accordance with other studies showing that the combination of HbA1c and FPG yields higher sensitivity and specificity for detecting DM [69, 73, 74]. This is also in line with current European guidelines, stating that screening for DM in patients with established CAD is initiated with the combination of HbA1c and FPG, and that an OGTT is added if the two tests are inconclusive [40].

In our study we found that FPG reproduced sufficiently after 4-6 weeks. The difference between in-hospital FPG and FPG at follow-up was not significant (p-value 0.066). However, due to a small sample size, this might be a false negative result. FPG is known to have a considerably intraindividual and preanalytical variability [43]. For instance, FPG measured early after an acute coronary syndrome may be influenced by stress-hyperglycaemia being associated with acute illness [75]. This would also influence an OGTT. Since HbA1c reflects the long-term glycaemic exposure, most factors that might influence plasma glucose measurements have little effect on the HbA1c concentration [43]. Thus, the combination of HbA1c and FPG might add diagnostic strength by adjusting for the biological and the preanalytical variability of FPG.

There are discrepancies in different recommendations regarding which levels of FPG and HbA1c that defines a person to be at high risk of developing DM [2, 4, 76]. The only method on which there is an agreement regarding the definition of ‘high risk’ is the OGTT, i.e. IGT. Therefore, further research on the clinical relevance of high-risk groups defined by FPG and HbA1c is needed before HbA1c and FPG can replace the OGTT in identification of people at high risk of future DM. Current guidelines still recommend that identification of individuals at high risk of future DM (i.e. IGT) should be done by performing an OGTT [40].

4.1 Clinical relevance

We evaluated HbA1c- and FPG measurements compared to an OGTT, in a subgroup of patients included in the Gluko-NorStent study. Our results may be of value when potential associations between dysglycaemia and outcome in the large NorStent trial are to be analysed.

4.2 Limitations

Our study is limited by a small sample size. Furthermore, the clinical diagnosis regarding stable or unstable coronary artery disease was not available for this student project. Thus, our finding may be biased by the fact that results may differ between patients with stable CAD versus acute coronary syndrome.

4.3 Conclusion

Our study demonstrated that HbA1c $\geq$ 6.5 % underdiagnosed DM compared with an OGTT, while HbA1c values in the range 5.7-6.4 % seemed to overestimate the risk of having IGT. There was a poor association between in-hospital HbA1c values $\geq$ 5.7 % and abnormal glucose tolerance defined by an OGTT performed 4-6 weeks following PCI. An HbA1c $\geq$ 6.0 % performed better at identifying patients with abnormal glucose regulation diagnosed by an OGTT. The combination of HbA1c and FPG added diagnostic strength compared to HbA1c in terms of identifying abnormal glucose regulation confirmed by an OGTT. Finally,FPG measured during hospitalization for PCI treatment reproduced sufficiently after 4-6 weeks.

Our findings support current European guidelines on detection of abnormal glucose regulation in patients with established cardiovascular disease, stating that identification of DM should be initiated with the combination of FPG and HbA1c, followed by an OGTT if the two tests are inconclusive. With respect to identification of patients at risk of DM, i.e. IGT, an OGTT should be performed.

5 Acknowledgements

We thank Professor Vibeke Videm for help with statistical analyses and we thank the research nurses Ann Mari Myraunet and Tove Vindsetmo for carefully administrating and performing the oral glucose tolerance tests being the basis for this project.

6 References

1. *Definition and diagnosis of diabetes mellitus and intermediate hyperglycaemia. Report of WHO/IDF consultation. Geneva: World Health Organization, International Diabetes Federation.* 2006.
2. *Diagnosis and classification of diabetes mellitus.* Diabetes Care, 2010. **33 Suppl 1**: p. S62-9.
3. *WHO Guidelines Approved by the Guidelines Review Committee*, in *Use of Glycated Haemoglobin (HbA1c) in the Diagnosis of Diabetes Mellitus: Abbreviated Report of a WHO Consultation*. 2011, World Health Organization

4. *International Expert Committee report on the role of the A1C assay in the diagnosis of diabetes.* Diabetes Care, 2009. **32**(7): p. 1327-34.
5. Hanssen, K.F., et al., *Blood glucose control and diabetic microvascular complications: long-term effects of near-normoglycaemia.* Diabet Med, 1992. **9**(8): p. 697-705.
6. Fox, C.S., et al., *Increasing cardiovascular disease burden due to diabetes mellitus: the Framingham Heart Study.* Circulation, 2007. **115**(12): p. 1544-50.
7. *Classification and diagnosis of diabetes mellitus and other categories of glucose intolerance. National Diabetes Data Group.* Diabetes, 1979. **28**(12): p. 1039-57.
8. Rushforth, N.B., M. Miller, and P.H. Bennett, *Fasting and two-hour post-load glucose levels for the diagnosis of diabetes. The relationship between glucose levels and complications of diabetes in the Pima Indians.* Diabetologia, 1979. **16**(6): p. 373-9.
9. *Report of the Expert Committee on the Diagnosis and Classification of Diabetes Mellitus.* Diabetes Care, 1997. **20**(7): p. 1183-97.
10. Organization, W.H., *Use of glycated haemoglobin (HbA1c) in the diagnosis of diabetes mellitus: abbreviated report of WHO consultation*. 2011: Geneva
11. *International Diabetes Federation. IDF Diabetes Atlas update poster, 6th edn. Brussels, Belgium: International Diabetes Federation, 2014.*
12. Guariguata, L., et al., *Global estimates of diabetes prevalence for 2013 and projections for 2035.* Diabetes Res Clin Pract, 2014. **103**(2): p. 137-49.
13. Goldschmidt-Clermont, P.J., et al., *Atherosclerosis, inflammation, genetics, and stem cells: 2012 update.* Curr Atheroscler Rep, 2012. **14**(3): p. 201-10.
14. Weber, C. and H. Noels, *Atherosclerosis: current pathogenesis and therapeutic options.* Nat Med, 2011. **17**(11): p. 1410-22.
15. Grundy, S.M., *Pre-diabetes, metabolic syndrome, and cardiovascular risk.* J Am Coll Cardiol, 2012. **59**(7): p. 635-43.
16. Paneni, F., et al., *Diabetes and vascular disease: pathophysiology, clinical consequences, and medical therapy: part I.* Eur Heart J, 2013. **34**(31): p. 2436-43.
17. Biondi-Zoccai, G.G., et al., *Atherothrombosis, inflammation, and diabetes.* J Am Coll Cardiol, 2003. **41**(7): p. 1071-7.
18. Hegab, Z., et al., *Role of advanced glycation end products in cardiovascular disease.* World J Cardiol, 2012. **4**(4): p. 90-102.
19. Kip, K.E., et al., *Coronary angioplasty in diabetic patients. The National Heart, Lung, and Blood Institute Percutaneous Transluminal Coronary Angioplasty Registry.* Circulation, 1996. **94**(8): p. 1818-25.

20. Kurihara, O., et al., *Coronary atherosclerosis is already ongoing in pre-diabetic status: Insight from intravascular imaging modalities.* World J Diabetes, 2015. **6**(1): p. 184-91.
21. Ando, H., et al., *Comparison of tissue characteristics between acute coronary syndrome and stable angina pectoris. An integrated backscatter intravascular ultrasound analysis of culprit and non-culprit lesions.* Circ J, 2011. **75**(2): p. 383-90.
22. Boden, W.E., et al., *Optimal medical therapy with or without PCI for stable coronary disease.* N Engl J Med, 2007. **356**(15): p. 1503-16.
23. Frye, R.L., et al., *A randomized trial of therapies for type 2 diabetes and coronary artery disease.* N Engl J Med, 2009. **360**(24): p. 2503-15.
24. Windecker, S., et al., *2014 ESC/EACTS Guidelines on myocardial revascularization: The Task Force on Myocardial Revascularization of the European Society of Cardiology (ESC) and the European Association for Cardio-Thoracic Surgery (EACTS)Developed with the special contribution of the European Association of Percutaneous Cardiovascular Interventions (EAPCI).* Eur Heart J, 2014. **35**(37): p. 2541-619.
25. Vinik, A.I., et al., *Diabetic autonomic neuropathy.* Diabetes Care, 2003. **26**(5): p. 1553-79.
26. *The final 10-year follow-up results from the BARI randomized trial.* J Am Coll Cardiol, 2007. **49**(15): p. 1600-6.
27. Kapur, A., et al., *Randomized comparison of percutaneous coronary intervention with coronary artery bypass grafting in diabetic patients. 1-year results of the CARDia (Coronary Artery Revascularization in Diabetes) trial.* J Am Coll Cardiol, 2010. **55**(5): p. 432-40.
28. Kappetein, A.P., et al., *Treatment of complex coronary artery disease in patients with diabetes: 5-year results comparing outcomes of bypass surgery and percutaneous coronary intervention in the SYNTAX trial.* Eur J Cardiothorac Surg, 2013. **43**(5): p. 1006-13.
29. Farkouh, M.E., et al., *Strategies for multivessel revascularization in patients with diabetes.* N Engl J Med, 2012. **367**(25): p. 2375-84.
30. StephanWindecker, et al., *2014 ESC/EACTS guidelines on myocardial revascularization.* Rev Esp Cardiol (Engl Ed), 2015. **68**(2): p. 144.
31. Harskamp, R.E. and D.W. Park, *Percutaneous coronary intervention in diabetic patients: should choice of stents be influenced?* Expert Rev Cardiovasc Ther, 2013. **11**(5): p. 541-53.
32. Mehran, R., et al., *Short- and long-term results after multivessel stenting in diabetic patients.* J Am Coll Cardiol, 2004. **43**(8): p. 1348-54.
33. Hoffmann, R., et al., *Patterns and mechanisms of in-stent restenosis. A serial intravascular ultrasound study.* Circulation, 1996. **94**(6): p. 1247-54.
34. Aronson, D. and E.R. Edelman, *Revascularization for coronary artery disease in diabetes mellitus: angioplasty, stents and coronary artery bypass grafting.* Rev Endocr Metab Disord, 2010. **11**(1): p. 75-86.
35. Morice, M.C., et al., *A randomized comparison of a sirolimus-eluting stent with a standard stent for coronary revascularization.* N Engl J Med, 2002. **346**(23): p. 1773-80.
36. Bangalore, S., et al., *Outcomes with various drug eluting or bare metal stents in patients with diabetes mellitus: mixed treatment comparison analysis of 22,844 patient years of follow-up from randomised trials.* Bmj, 2012. **345**: p. e5170.

37. Sheiban, I., et al., *Impact of diabetes mellitus on early and long-term results of percutaneous drug-eluting stent implantation for unprotected left main coronary disease.* J Cardiovasc Med (Hagerstown), 2008. **9**(12): p. 1246-53.
38. Jimenez-Quevedo, P., et al., *Long-term clinical benefit of sirolimus-eluting stent implantation in diabetic patients with de novo coronary stenoses: long-term results of the DIABETES trial.* Eur Heart J, 2007. **28**(16): p. 1946-52.
39. Baumgart, D., et al., *One-year results of the SCORPIUS study: a German multicenter investigation on the effectiveness of sirolimus-eluting stents in diabetic patients.* J Am Coll Cardiol, 2007. **50**(17): p. 1627-34.
40. Ryden, L., et al., *ESC Guidelines on diabetes, pre-diabetes, and cardiovascular diseases developed in collaboration with the EASD: the Task Force on diabetes, pre-diabetes, and cardiovascular diseases of the European Society of Cardiology (ESC) and developed in collaboration with the European Association for the Study of Diabetes (EASD).* Eur Heart J, 2013. **34**(39): p. 3035-87.
41. Wiemer, M., et al., *Drug-eluting stents with biodegradable polymer for the treatment of patients with diabetes mellitus: clinical outcome at 2 years in a large population of patients.* Med Devices (Auckl), 2015. **8**: p. 153-60.
42. Bartoli, E., G.P. Fra, and G.P. Carnevale Schianca, *The oral glucose tolerance test (OGTT) revisited.* Eur J Intern Med, 2011. **22**(1): p. 8-12.
43. Sacks, D.B., *A1C versus glucose testing: a comparison.* Diabetes Care, 2011. **34**(2): p. 518-23.
44. Tessier, F.J., *The Maillard reaction in the human body. The main discoveries and factors that affect glycation.* Pathologie Biologie, 2010. **58**(3): p. 214-219.
45. Nathan, D.M., H. Turgeon, and S. Regan, *Relationship between glycated haemoglobin levels and mean glucose levels over time.* Diabetologia, 2007. **50**(11): p. 2239-44.
46. Klein, R., et al., *Glycosylated hemoglobin predicts the incidence and progression of diabetic retinopathy.* Jama, 1988. **260**(19): p. 2864-71.
47. Gallagher, E.J., D. Le Roith, and Z. Bloomgarden, *Review of hemoglobin A(1c) in the management of diabetes.* J Diabetes, 2009. **1**(1): p. 9-17.
48. Pani, L.N., et al., *Effect of aging on A1C levels in individuals without diabetes: evidence from the Framingham Offspring Study and the National Health and Nutrition Examination Survey 2001-2004.* Diabetes Care, 2008. **31**(10): p. 1991-6.
49. Herman, W.H. and R.M. Cohen, *Racial and ethnic differences in the relationship between HbA1c and blood glucose: implications for the diagnosis of diabetes.* J Clin Endocrinol Metab, 2012. **97**(4): p. 1067-72.
50. *HbA1c as a predictor of diabetes and as an outcome in the diabetes prevention program: a randomized clinical trial.* Diabetes Care, 2015. **38**(1): p. 51-8.
51. Pajunen, P., et al., *HbA(1c) in diagnosing and predicting Type 2 diabetes in impaired glucose tolerance: the Finnish Diabetes Prevention Study.* Diabet Med, 2011. **28**(1): p. 36-42.
52. Ramachandran, A., et al., *Predictive value of HbA1c for incident diabetes among subjects with impaired glucose tolerance--analysis of the Indian Diabetes Prevention Programmes.* Diabet Med, 2012. **29**(1): p. 94-8.
53. Pinelli, N.R., et al., *Sensitivity and specificity of glycated hemoglobin as a diagnostic test for diabetes and prediabetes in Arabs.* J Clin Endocrinol Metab, 2011. **96**(10): p. E1680-3.
54. van 't Riet, E., et al., *Relationship between A1C and glucose levels in the general Dutch population: the new Hoorn study.* Diabetes Care, 2010. **33**(1): p. 61-6.
55. Buell, C., D. Kermah, and M.B. Davidson, *Utility of A1C for diabetes screening in the 1999 2004 NHANES population.* Diabetes Care, 2007. **30**(9): p. 2233-5.

56. Rohlfing, C.L., et al., *Use of GHb (HbA1c) in screening for undiagnosed diabetes in the U.S. population.* Diabetes Care, 2000. **23**(2): p. 187-91.
57. Carson, A.P., et al., *Comparison of A1C and fasting glucose criteria to diagnose diabetes among U.S. adults.* Diabetes Care, 2010. **33**(1): p. 95-7.
58. Kramer, C.K., M.R. Araneta, and E. Barrett-Connor, *A1C and diabetes diagnosis: The Rancho Bernardo Study.* Diabetes Care, 2010. **33**(1): p. 101-3.
59. *Trial of Drug Eluting Stent Versus Bare Metal Stent to Treat Coronary Artery Stenosis (NORSTENT)* Available from: https://clinicaltrials.gov/ct2/show/NCT00811772?term=norstent&rank=1
60. Alberti, K.G. and P.Z. Zimmet, *Definition, diagnosis and classification of diabetes mellitus and its complications. Part 1: diagnosis and classification of diabetes mellitus provisional report of a WHO consultation.* Diabet Med, 1998. **15**(7): p. 539-53.
61. Hage, C., et al., *Fasting glucose, HbA1c, or oral glucose tolerance testing for the detection of glucose abnormalities in patients with acute coronary syndromes.* Eur J Prev Cardiol, 2013. **20**(4): p. 549-54.
62. Farhan, S., et al., *Comparison of HbA1c and oral glucose tolerance test for diagnosis of diabetes in patients with coronary artery disease.* Clin Res Cardiol, 2012. **101**(8): p. 625-30.
63. de Mulder, M., et al., *Comparison of diagnostic criteria to detect undiagnosed diabetes in hyperglycaemic patients with acute coronary syndrome.* Heart, 2012. **98**(1): p. 37-41.
64. Gyberg, V., et al., *Screening for dysglycaemia in patients with coronary artery disease as reflected by fasting glucose, oral glucose tolerance test, and HbA1c: a report from EUROASPIRE IV--a survey from the European Society of Cardiology.* Eur Heart J, 2015. **36**(19): p. 1171-7.
65. Donahoe, S.M., et al., *Diabetes and mortality following acute coronary syndromes.* Jama, 2007. **298**(7): p. 765-75.
66. Qiao, Q., et al., *Two prospective studies found that elevated 2-hr glucose predicted male mortality independent of fasting glucose and HbA1c.* J Clin Epidemiol, 2004. **57**(6): p. 590-6.
67. Meigs, J.B., et al., *Fasting and postchallenge glycemia and cardiovascular disease risk: the Framingham Offspring Study.* Diabetes Care, 2002. **25**(10): p. 1845-50.
68. Lorenzo, C., et al., *A1C between 5.7 and 6.4% as a marker for identifying pre-diabetes, insulin sensitivity and secretion, and cardiovascular risk factors: the Insulin Resistance Atherosclerosis Study (IRAS).* Diabetes Care, 2010. **33**(9): p. 2104-9.
69. Hu, Y., et al., *Combined use of fasting plasma glucose and glycated hemoglobin A1c in the screening of diabetes and impaired glucose tolerance.* Acta Diabetol, 2010. **47**(3): p. 231-6.
70. *Is fasting glucose sufficient to define diabetes? Epidemiological data from 20 European studies. The DECODE-study group. European Diabetes Epidemiology Group. Diabetes Epidemiology: Collaborative analysis of Diagnostic Criteria in Europe.* Diabetologia, 1999. **42**(6): p. 647-54.
71. Bartnik, M., et al., *Oral glucose tolerance test is needed for appropriate classification of glucose regulation in patients with coronary artery disease: a report from the Euro Heart Survey on Diabetes and the Heart.* Heart, 2007. **93**(1): p. 72-7.
72. Unwin, N., et al., *Impaired glucose tolerance and impaired fasting glycaemia: the current status on definition and intervention.* Diabet Med, 2002. **19**(9): p. 708-23.
73. Tekumit, H., et al., *Diagnostic value of hemoglobin A1c and fasting plasma glucose levels in coronary artery bypass grafting patients with undiagnosed diabetes mellitus.* Ann Thorac Surg, 2010. **89**(5): p. 1482-7.

74. Perry, R.C., et al., *HbA1c measurement improves the detection of type 2 diabetes in high-risk individuals with nondiagnostic levels of fasting plasma glucose: the Early Diabetes Intervention Program (EDIP).* Diabetes Care, 2001. **24**(3): p. 465-71.
75. Dungan, K.M., S.S. Braithwaite, and J.C. Preiser, *Stress hyperglycaemia.* Lancet, 2009. **373**(9677): p. 1798-807.
76. Olson, D.E., et al., *Screening for diabetes and pre-diabetes with proposed A1C-based diagnostic criteria.* Diabetes Care, 2010. **33**(10): p. 2184-9.