Summary
Type 2 diabetes is a chronic disorder characterized by hyperglycemia (elevated glucose levels), with an increasing worldwide prevalence and a high risk of complications like cardiovascular disease. Many risk factors for type 2 diabetes have been identified so far. However, since type 2 diabetes is a multifactorial disease, questions remain on combining risk factors in the prediction of type 2 diabetes, the interaction between several risk factors in the occurrence of type 2 diabetes and the role of genetic factors in type 2 diabetes risk. In this thesis, we investigated the prevalence of type 2 diabetes and intermediate hyperglycemia (slightly elevated glucose levels) in the Netherlands in 1989 and 2006, and examined adiposity, genetic factors and family history of diabetes as risk factors for type 2 diabetes, using data from the Hoorn Study, the New Hoorn Study and the Nurses’ Health Study. In addition, we focused on the three markers of hyperglycemia: fasting glucose, postload glucose (glucose levels 2 hours after a standard 75gram oral glucose tolerance test (OGTT)) and glycated hemoglobin (HbA1c). Fasting glucose and postload glucose are used for the diagnosis of type 2 diabetes, while HbA1c is used to monitor glycemic control in type 2 diabetes patients. Recently, it was proposed to use HbA1c levels ≥ 6.5% for the diagnosis of diabetes instead of elevated glucose levels, resulting in an ongoing discussion whether or not to adapt this advice. In line with this, we explored the agreement between the three different markers of hyperglycemia, and investigated the use of HbA1c for the diagnosis of type 2 diabetes. Last, we investigated the association of the three markers of hyperglycemia with the development of cardiovascular morbidity and mortality.

The Hoorn Study is a prospective cohort study, which started in 1989 among 2484 randomly selected Caucasian residents of the town of Hoorn, the Netherlands, between 50 and 75 years of age. In 2006, the New Hoorn Study was initiated, in which 2807 Caucasian residents of Hoorn between 40 and 65 years of age participated. In both samples, fasting glucose was determined and an OGGT was performed. Fasting glucose and postload glucose were measured.
Summary

Glucose were used to categorize participants of the Hoorn Study and New Hoorn Study in groups of glucose metabolism (according to the 2006 criteria of the World Health Organization): normal glucose metabolism, intermediate hyperglycemia and type 2 diabetes. A comparison of the two samples revealed that between 1989 and 2006, the prevalence of intermediate hyperglycemia increased significantly from 14.4% to 17.6% in the overlapping age category of participants between 50 and 65 years of age. In addition, the prevalence of type 2 diabetes remained stable at 8%. Improvements in physical activity and lipid profile were observed, however, the prevalence of adiposity and hypertension increased (Chapter 2).

One of the strongest risk factors for type 2 diabetes is adiposity. Still, 83.7% of the participants of the Hoorn Study with adiposity at baseline did not develop type 2 diabetes after 6 years of follow-up. Therefore, the prediction of type 2 diabetes should not be based on adiposity solely. The use of a diabetes risk score, in which risk factors for type 2 diabetes are combined, can be of help to identify those at risk of developing type 2 diabetes in the near future (Chapter 3). One of these other risk factors is a family history of diabetes. In the Nurses’ Health Study, a prospective cohort study started in 1979 among 121,700 female nurses in the United States, a family history of diabetes was associated with a twofold increased risk of incident type 2 diabetes after 20 years of follow-up (Chapter 4). It is often suggested that this strong association is the result of genetic influences. However, the clustering of body fatness in families, which can be due to both shared genetic and shared environmental influences, may also contribute to the relationship between family history of diabetes and the development of type 2 diabetes. In the Nurses’ Health Study, we explored the role of adiposity in this association between family history of diabetes and incident type 2 diabetes. This revealed that 21% of the association between family history of diabetes and incident type 2 diabetes could be explained by adiposity (Chapter 4).
Due to the increase in genome-wide association (GWA) studies, evidence on the role of genetic factors in the development of type 2 diabetes is emerging. Specific genetic locations (loci's) in relation to type 2 diabetes and glucose levels have been determined. To increase susceptibility for a true genetic association, results from GWA studies need to be replicated in different populations. We replicated the association of 4 genetic locations [glucokinase (GCK), glucokinase regulatory protein (GCKR), islet-specific glucose 6 phosphatase catalytic subunit-related protein (G6PC2) and melatonin receptor type 1B (MTNR1B)] with fasting glucose, and studied the association of these 4 loci’s with susceptibility to type 2 diabetes and level of HbA1c. In this case-control study among 2628 patients with type 2 diabetes (cases) and 2041 normoglycemic individuals (control), significant associations of GCK, G6PC2 and MTNR1B with fasting glucose were found, as well as significant associations of GCK and G6PC2 with HbA1c. The contribution of each individual genetic variant to the risk of type 2 diabetes was found to be very low. However, when calculating a sum score, in which the risk alleles in the genes for GCK, GCKR, G6PC2 and MTNR1B were combined, revealed a significant association with type 2 diabetes, levels of fasting glucose and HbA1c, indicating that genetic variants have a combined effect on type 2 diabetes risk (Chapter 5).

Next to our research into the prevalence and risk factors for hyperglycemia, we investigated three markers of hyperglycemia: fasting glucose, postload glucose and HbA1c. In the population of the New Hoorn Study, the correlations between fasting glucose and HbA1c, and between postload glucose and HbA1c were low in people without diabetes, 0.46 and 0.33 respectively. In type 2 diabetes patients, these correlations were 0.71 and 0.79 respectively. In addition, we investigated the diagnostic properties of HbA1c for the diagnosis of type 2 diabetes, using the OGTT as gold standard. The optimal cut-off point for the diagnosis of type 2 diabetes based on HbA1c in the population of the New Hoorn Study, was an HbA1c level of 5.8%, displaying a sensitivity of
Summary

72% and a specificity of 91%. The officially proposed cut-off point of HbA1c of 6.5% for the diagnosis of type 2 diabetes, had low diagnostic properties as compared to the use of an OGTT, with a sensitivity of 24% (Chapter 6).

In the Hoorn Study population, the baseline measurements in 1989 are extended with follow-up measurements in 1996 and 2000. This created the opportunity to study developmental patterns of glucose and HbA1c in those who never developed type 2 diabetes during follow-up, compared to those who developed type 2 diabetes. The results showed that in those who were diagnosed with type 2 diabetes in 2000, but who were free of type 2 diabetes in 1989 and 1996, glucose levels increase slightly, but non-significantly, between 1989 and 1996, followed by a rapid and significant increase between 1996 and 2000. HbA1c levels already displayed a significant increase between 1989 and 1996, also followed by a rapid increase between 1996 and 2000. These results implicate that HbA1c and glucose might show different developmental patterns before the diagnosis of type 2 diabetes, characterized by an early increase in HbA1c, followed by rapid increase in HbA1c and glucose shortly before diagnosis (Chapter 7).

A continuous registration of morbidity and mortality of the participants of the Hoorn Study was used to investigate the association of HbA1c and glucose with 10-year incidence of fatal and non-fatal cardiovascular disease in 1647 participants without type 2 diabetes at baseline. Adjusted for traditional CVD risk factors, high levels of HbA1c were found to be significantly related to the risk of developing non-fatal CVD, especially in women, with a 2.3 higher risk of developing non-fatal CVD in women in the upper 10% of the range of HbA1c levels (HbA1c ≥ 6.0%) as compared to women in the lowest quartile of HbA1c levels (HbA1c ≤ 5.1%). Associations between HbA1c and incident CVD events in men, as well as between glucose, either fasting or postload, and incident CVD events in both sexes, were explained by traditional CVD risk factors.
In the general discussion (Chapter 9), several issues were discussed. Firstly, we addressed some methodological aspects in relation to the studies included in this thesis, mainly focussing on possible sources of bias and factors that might harm the precision of our results presented. Secondly, we discussed the complexity of the evolvement of type 2 diabetes. Many risk factors for type 2 diabetes have been identified, but the presence of all of these risk factors together does not guarantee a 100% likelihood of developing type 2 diabetes in the future. Therefore, the prediction of future diabetes remains a challenge. The use of a type 2 diabetes risk score, which includes several diabetes risk factors, might be helpful in identifying those at high risk of future type 2 diabetes. More research, including gene-gene and gene-environment interaction studies are needed to further investigate the occurrence of type 2 diabetes. Last, we discussed some explanations for the lack of concordance between the three markers of hyperglycemia found in this thesis. Several studies have provided evidence on the role of non-glycemic factors in the determination of HbA1c, including for example genetic factors, age and intra-individual differences in erythrocyte environment. In addition, we observed limited diagnostic abilities of HbA1c as compared the OGTT for the diagnosis of diabetes. Moreover, little is known about the effect of starting a diabetes treatment in individuals with HbA1c levels ≥ 6.5%, but normal glucose levels. As a result, the use of HbA1c for the diagnosis of diabetes instead of elevated glucose levels should be considered with caution. More research on consequences of the use of HbA1c for the diagnosis of diabetes is needed before the advice to introduce HbA1c for the worldwide diagnosis of diabetes is implemented.