Chapter 7

Changes in HbA1c level and adjusting glucose-lowering medication in long term follow up of T2D, a population-based study in the Netherlands

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ABSTRACT

BACKGROUND Long term glycemic control has been a struggle in trading type 2 diabetes (T2D) patients for decades, being up 60% of patient may not achieve it in long term. The current historical cohort analysis of a real-world medicare registry data was conducted to assess and compare the effect of glucose-lowering medication changes/intensifying on HbA1c level over long term among T2D patients.

METHODS We used a long-term follow-up data of frequently monitoring HbA1c level over the past 25 years (1985-2010) of 570 patients with T2D from a routine primary care setting in the Netherlands being representative. Four therapeutic agents roof Metformin (MET), Sulfonylurea (SU) Thiazolidine(TZD) and Insulin (INS), according ATC classification. The prescribed and equivalents dose of medication was extracted and the total dose was expressed as the number of tablets used daily, generating the number of equivalents used by the patients. We modeled subject-specific temporal variation in HbA1c concentration as a function of the number of years since the initial measurement. We fitted subject-specific Gaussian generalized additive models when we calculated the derivatives, stated as rate of change in HbA1c in % per year, for each individual using linear mixed models. Fixed effects are included up to two-way interactions between binary indicator variables for each of the 4 medical treatments (MET, SU, TZD, INS) and additional additive effects of age at initial measurement (standardized) and sex (male = 0, 1).

RESULTS Insulin was associated with a reduction in HbA1c at 9.8% relative to non-Insulin. HbA1c levels in presence of other factors increased significantly by increasing SU up to 0.045 units (P=0.003), and did not change by increasing TZD (P=0.2167), whereas decreased 0.0297 unit by increasing MET which was not significant (P=0.056). When we included combination therapies into model, HbA1c decreased significantly 0.093 units by combination of MET and SU (P<0.001), 0.358 units by combination of INS and TZD (P=0.0009) and 0.169 units by combination of SU and TZD (P=0.0014). Effect of MET, TZD, or INS plus SU were not significant in this model. This may indicate that there was no significant difference in HbA1c values by different levels of these medications over the follow-up time.

CONCLUSION The initial monotherapy with either MET or TZD alone or even a combination of INS with MET, or INS with SU fails to achieve a low HbA1c level in long term. INS remains an important part of treatment for T2D, either alone, or in combination with TZD. To prohibit development of INS resistance, a combination of TZD with MET or SU seems as the most effective alternative in reducing HbA1c.
INTRODUCTION

Diabetes is expected to affect 552 million people in 2030 worldwide being the majority of type 2 diabetes (T2D) especially in developing countries\(^1\). T2D is caused by a pathogenic defect in the pancreases’ beta-cells and Insulin (INS) resistance throughout the body that yields hyperglycemia\(^2\). The goal of all treatment strategies for T2D is to maintain blood glucose concentrations to an approximately normal range to reduce the effects of chronic hyperglycemia, while avoiding serious hypoglycemic events which often are not achieved by current therapies. That leads to uncontrolled hyperglycemia which is associated with an increase in the risk of microvascular complications of T2D such as blindness, renal failure, cardiovascular diseases (CVD), and early mortality\(^3\). Eventually, T2D and its associated health complications are one of the most common non-communicable and increasing global health problems for all age groups worldwide\(^4\). There is evidence that tight glucose control, especially in the early years after diagnosis, reduces the risk of long-term CVD complications in patients with T2D\(^5\). A commonly used measure of average blood glucose control is measuring glycated hemoglobin (HbA1c), which reflects plasma glucose control over the past two to three months. HbA1c has been shown to be strongly related to the occurrence of complications in T2D patients in prospective observational studies\(^6\). The target level of HbA1c was recommended with a variation of levels as 6.5-7.5% in NICE guidance and <7.0% by the American Diabetes Association (ADA)\(^7\). This is expected to be achieved by changing to a more physically active lifestyle and using hypoglycemic agents\(^8\). Strict glycemic control is the most important factor in managing T2D patients but with different strategies for different patients.

T2D patients are often treated with more than one drug, including oral antidiabetic drugs (OAD) and drugs used to treat diabetic complications, such as dyslipidemia and hypertension. Pharmacologically, T2D is treated with nine major classes of oral and injectable approved drugs. Oral anti-diabetic drugs are among the most widely prescribed.

Recent observational studies and randomized controlled trials have mainly shown broadly similar reductions in HbA1c by the anti-hyperglycemic agents when used as initial monotherapy rather than as add-on treatments in patients with HbA1c of approximately 9.0%\(^9\). Metformin (MET) has been known as a usual first-line therapy when diet and exercise are insufficient. For most patients in who MET alone is no longer sufficient, the options include adding a Sulphonylurea (SU), or a Thiazolidinedione (TZD)\(^10\). Met and SU are the most commonly prescribed oral anti-hyperglycemic medication for achieving glycemic control for
a lower level baseline of HbA1c, but not effective for patients with a baseline of HbA1c ≥9%. Monotherapy with MET or SU has failing range of up to 50% to achieve the target level of HbA1c <7%. INS replacement therapy is recommended and preferred prescribed when patients are not successful in maintaining their blood glucose levels or unwanted side effects through OAD agents. Due to the complexity of INS treatment and also patients' heterogeneity, the decision to switch to INS is made based on patients' individual needs as well as physicians' judgments. International guidelines, therefore, recommend an individualized treatment strategy to achieve and maintain target levels of glycemic control. Due to the progressive decline in beta-cells function many patients fail to maintain an almost nearly normal HbA1c range. Therefore, the ADA and the European Association for the Study of Diabetes (EASD) recommend early initiation of basal INS with its effect on beta-cell function the most effective therapy especially for patients that have failed for achieving or maintaining HbA1c goal (<6.5 -7.0%) with monotherapy by OAD.

The pattern of combination therapy of anti-hyperglycemic treatment and their relationship with HbA1c level changes over long-term follow up after diagnosis of T2D in most of the studies is unknown. Other studies have conducted in a centrally organized care system that is not representative of routine diabetes care and differs from the routine primary care system. Thus the current historical cohort analysis of real-world medicare registry data was conducted to assess and compare the effect of glucose-lowering medication changes/intensifying and demographic characteristics on HbA1c level over time among T2D patients with opportunity of using a long-term follow-up data of frequently monitoring HbA1c level over the past 25 years. This cohort of patients with T2D from a routine primary care setting in the Netherlands may be representative of routine primary care practice and the results are therefore directly relevant for daily clinical practice.

MATERIALS and METHODS
Study design and data source

In this study, with real-world medicare registry, the HbA1c measurements were more unequally scheduled over time, and in addition, there were differences in the timings of OAD and INS initiation. The study was conducted within the framework of the Utrecht Cardiovascular Pharmacogenetic studies (UCP). UCP comprises subjects derived from the PHARMO Database Network, a population-based network of electronic healthcare databases (PHARMO, www.pharmo.nl). The PHARMO Database Network combines data from different primary and secondary healthcare settings in the Netherlands. For this study, data from the Hospitalisation Database linked to the Out-patient Pharmacy Database were used.
At the time of the study, the base population covered approximately 2,000,000 community-dwelling inhabitants of several population-defined areas in the Netherlands. This database has been extensively described and validated and has been used previously in over 20 studies. This study was conducted in a cohort of patients diagnosed with T2D obtained from Dutch routine primary care between 1985 and 2010 to determine the efficacy of anti-hyperglycemic agents on HbA1c level. Approval for this study was obtained from the Medical Ethics Committee of the University Medical Center Utrecht, The Netherlands. Participants were recruited through community pharmacies, where they received a letter in which the purpose of the study was explained. They were asked to return an informed consent form and a filled-out questionnaire. All participants were explicitly asked to consent for the collection, storage and genotyping of the DNA material.

Population

The source population consisted of T2D patients from UCP to study T2D treatment. The diagnosis of T2D was defined via the use of at least one anti-diabetic drug either oral hypoglycemic agents or INS in combination with oral or as the second line for treatment based on pharmacy records. This longitudinal database contains data extracted from the electronic medical records of patients with T2D managed by routine general practice. This database includes prescription data, medical histories, results from routine laboratory tests for over 570 patients diagnosed with T2D.

Inclusion criteria

Patients were included if they met the criteria for inclusion by being 18 years or older and at least three HbA1c measurements in the follow-up period.

Outcomes

The HbA1c level was defined as the outcome of interest and was evaluated as a proxy to estimate the cumulative effect of using anti-hyperglycemic agent during the follow-up time. HbA1c was measured using standard protocols and was normalized according to local units.

Medication

Four therapeutic agent regimes were classified into two major therapeutic groups including oral (MET, SU, TZD) and injectable (INS). The OAD medication was clas-
sified based on the ATC class. The prescribed and equivalents dose of OAD medication was generated based on the number of daily used tablets during the period of four months prior and after the measurement of HBA1c for the patients. The total dose was expressed as the number of tablets used daily, generating the number of equivalents used by the patients. A data matrix was created for the HbA1c for all patients having three or more measurements and using antidiabetic agents. The treatment status as INS or OAD total dose in the period of 120 days prior the measurement date of HbA1c and 120 days after was included in the matrix data. Demographic information was attached from the patients files.

**Statistical analysis**

First we modeled subject-specific temporal variation in HbA1c concentration as a function of the number of years since the initial measurement. We fitted subject-specific Gaussian generalized additive models (gams) with R version 3.6.0\(^1\) and RStudio version 1.2.1114\(^2\), using the gam function of the mgcv package version 1.8-28\(^3\). To fit individual-level “random” smooths, we used the bs= “fs” option of the gam function, which applies a shared smoothing penalty to all subjects. We used the m = 1 option in the s function to prevent over smoothing, which amounts to penalizing the squared first derivative of the smoother\(^4\). The number of basic functions was set at k = 20, and we used the gam.check function to verify its adequacy\(^5\). Parameters were estimated using restricted maximum likelihood (REML).

To estimate the direction and magnitude of temporal change in HbA1c concentration, we calculated the derivatives (rate of change in HbA1c in % per year) of the fitted smoothers for each individual, evaluated at each measurement point, using the fderiv function from the gratia package\(^6\) (Simpson 2019). Variation in derivatives was modeled using linear mixed models with the lmer function of the lme4 package\(^7\). We used individual ID and measurement block as random effects modifying model intercepts. As fixed effects we allowed for up to two-way interactions between binary indicator variables for each of the 4 medical treatments (MET, SU, TZD, INS) and additional additive effects of age at initial measurement (standardized) and sex (male = 0, 1). Significance of model parameters was assessed with Sattertwaite-corrected t-tests, using the lmerTest package\(^8\).

**RESULTS**

There was a wide distribution of age, ranging between 18- 88 years, being more men (64.6%) with mean age 57.9 (SD 11.3) than women with mean age 58.4 (SD 13.91) included.
We grouped the patients based on their prescribed medicine as 69.5% of patients only used oral (OAD: MET, SU, TZD) as monotherapy or in combination with other OAD, 30.5% of patients used combination therapy of OADs with INS injectable. We observed that 60.8% of patients developed at least one co-occurrence of CVD (30.4%), 17.01% had chronic eye disease and 13% had cancer. There were 2.3% of patients with neurologic disease and 1.3% with chronic kidney disease.

The aggregated longtime influences of these covariates on HbA1c levels are illustrated by Figure 1. It describes the overall changes in HbA1c levels and means observation carried forward data from the study against four main groups and combination of the prescribed medication. This figure shows the estimated HbA1c change over the 25-years period by baseline HbA1c levels and treatments. The number of HbA1c values included in this analysis was 9200. The mean HbA1c at the base line was 7.55 (SD 1.77) in the overall and was 7.55 (SD .77) at last time points with an overall coefficient (slope) of (SE .22).

Figure 2 shows adjusted mean HbA1c values for different treatments by time for a random set of five patients as an illustrative example per individual level data over follow up time. Per each patient we extracted available data time points which were analyzed. As illustrated the course of HBA1c varied per each patient over time from decreasing to increasing slops. The treatment-by-time interaction terms were not significant in these individually fitted models. Therefore, it may indicate that there were no significant differences in the changes over the time of HbA1c levels for the different treatments.
Next, an exposure–response relationship between the medication and HbA1c level in T2D was estimated using a mixed effect model. We fitted a series of step-wise multivariate model, to identify the most significant predictors. The complete list of variable estimates from the final exposure–response model is shown in Table 1. Among all tested covariates, INS and its combination with TZD, MET, SU and its combination with MET or TZD were found to have a significant effect on HbA1c levels. INS was associated with a reduction in HbA1c at 9.8% relative to non-INS. HbA1c levels in presence of other factors increased significantly by increasing SU up to 0.045 units (P=0.003), and did not change by increasing TZD (P=0.2167), whereas decreased by 0.0297 unit by increasing MET which was not significant (P=0.056). When we included combination therapies into model, HbA1c decreased significantly by 0.093 units by combination of MET and SU (P<0.001), 0.358 units by combination of INS and TZD (P=0.0009) and 0.169 units by combination of SU and TZD (P=0.0014). INS plus TZD therapeutic regiments led to a significantly less HbA1c levels, followed by SU plus TZD, and MET plus TZD. Effect of MET, TZD, or INS plus SU were not significant in this model. This may indicate that there were no significant differences in HbA1c values by different levels of these medications over the follow-up time.
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Table 1: Association of antihyperglycemic drug and HbA1c presented as % change in HbA1c per year.

<table>
<thead>
<tr>
<th>Predictor</th>
<th>Estimated effect (coefficients)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>INS</td>
<td>-0.098 ± 0.016</td>
<td>&lt;0.0001**</td>
</tr>
<tr>
<td>MET</td>
<td>-0.030 ± 0.016</td>
<td>0.0560*</td>
</tr>
<tr>
<td>SU</td>
<td>0.045 ± 0.015</td>
<td>0.0030**</td>
</tr>
<tr>
<td>TZD</td>
<td>0.064 ± 0.052</td>
<td>0.2167</td>
</tr>
<tr>
<td>INS*MET</td>
<td>-0.039 ± 0.023</td>
<td>0.0954</td>
</tr>
<tr>
<td>INS*SU</td>
<td>-0.034 ± 0.027</td>
<td>0.2012</td>
</tr>
<tr>
<td>INS*TZD</td>
<td>-0.360 ± 0.108</td>
<td>0.0009**</td>
</tr>
<tr>
<td>MET*SU</td>
<td>-0.093 ± 0.020</td>
<td>&lt;0.0001**</td>
</tr>
<tr>
<td>MET*TZD</td>
<td>-0.104 ± 0.050</td>
<td>0.0382*</td>
</tr>
<tr>
<td>SU*TZD</td>
<td>-0.170 ± 0.053</td>
<td>0.0014**</td>
</tr>
<tr>
<td>Age (z-score)</td>
<td>-0.010 ± 0.007</td>
<td>0.1668</td>
</tr>
<tr>
<td>Male</td>
<td>-0.027 ± 0.014</td>
<td>0.0840</td>
</tr>
</tbody>
</table>

# Linear mixed model with HbA1c as response variable, included as the derivative of fitted smoother, with a % change in HbA1c per year. P-values refer to t-tests with Satterthwaite's degrees of freedom. Estimated covariables include sex, age at the cohort entry and for all significant covariates and interactions in the final model are tested. Abbreviations: INS: Insulin, MET: Metformin, SU: Sulphonylurea, TZD: Thiazolidine. *shows interaction terms. SE: Standard error.

Figure 3: The coefficients of the linear mixed model.

The actual clinical HbA1c data (mean changes from baseline, obtained from a mixed model, with 95% CI) are shown as data points. Estimates of the main effects including INS, MET, SU and TZD are contrasts with the reference estimate obtained females with mean age that used no medication (that is estimate = 0.027 ± SE = 0.016 % per year, INS).

Figure 3 is a coefficient plot with 95% confidence intervals that represents interpretation of interaction terms. The actual clinical HbA1c data (mean change from baseline, obtained from a mixed model, with 95% CI) are shown as data points. Estimates of the main effects including INS, MET, SU and TZD are contrasts
with the reference estimate obtained females with mean age that used no med-
ication (that is estimate = 0.027±SE = 0.016 % per year, INS). Negative values
indicate decreasing slopes of fitted smoothers compared to reference value. In-
teraction terms indicate contrast with main effects, thus for example, combined
usage of INS plus TZD further reduced the slope by about 0.36±0.20 % per year.
SD ID and SD block indicate standard deviations of random intercepts between
individuals and blocks within individuals. The graph also shows the significance
interaction terms that are presented in Table 1. The adjusted mean HbA1c values
were significantly lower for combined INS and TZD in comparison with other OAD
over all-time points, but there was a different trajectory between OAD, and their
interaction with INS that are confirmed by the observed significant effect of inter-
action terms of these parameters.

**DISCUSSION**

We conducted a retrospective analysis for a large, population-based, observa-
tional cohort study. We utilized mixed effects models to determine the effect of
treatments associated with HbA1c as the most important predictor of the devel-
opment of complications, morbidity, and mortality of T2D. Using multi-step model
building process provided a final model with main and interaction term effect on
HbA1c level as the outcome of interest.

HbA1c levels in presence of other factors increased by increasing SU, whereas
decreased by increasing MET, INS and their combinations with TZD, SU and its
combination with MET or TZD. Treatment with combination of INS and TZD was
predicted to have more HbA1c reduction followed by INS, SU plus MET or TZD. INS
was also associated with a greater reduction in HbA1c comparing to non-INS. The
benefit of initiating an OAD agent is most apparent within the first 4 to 6 months,
with HbA1c levels unlikely to fall more than 1.5% on average. Pretreated HbA1c
levels have a modest effect on the fall of HbA1c levels in response to treatment.21
A recent systematic review suggested that patients with shorter diabetes duration
whom used initiating MET had lower HbA1c level at follow up.22

MET is one of the most popular oral glucose lowering medications, widely
considered to be the optimal initial therapy for patients with T2D. The prescrip-
tion’s efficacy has been demonstrated in monotherapy as well as in combination
with other glucose lowering medications for T2D. The findings of our prospective
observational study showed that MET was the most prescribed medication as a
first line using by 48% of patients during 25 years follow up. Other studies also
indicated that MET was recommended as the first-line in the treatment regime for
T2D23 because of decreasing blood glucose level without the risk of weight gain
and hypoglycemic effect as other OAD.24 However, our multivariate model of 25
years data showed that MET modestly decreased levels of HbA1c, but this effect just failed to reach statistical significance level.

There are too many studies that present achieving target level of < 7% for HbA1c ranged from 19% to 86% for highest to lowest baseline HbA1c in the first year after starting monotherapy by MET and long term follow up near 3 and 9 years showed ranging around 50% and 24% respectively for maintaining HbA1c lower than 7.0%\textsuperscript{25}. The same results were confirmed in another study with ranging from 14% to 81% for highest to lowest baseline HbA1c category respectively\textsuperscript{26}. Longer term follow up showed only 50% and 13% of patients were able to keep their HbA1c <7.0% at 3 and 9 years’ follow-up on their initial monotherapy by MET\textsuperscript{27}. Initial monotherapy with maximally tolerated dose of MET may not be sufficient for glycemic control with HbA1c range<6.5% and more frequently fails to maintain HbA1c < 7.0 %\textsuperscript{28}. Similar to those finding our study found a significant association for decreasing the HbA1c in patient whom used MET alone or in combination to other oral hypoglycemic agents. Dosage up MET is associated with 71% higher chance of attaining target HbA1c during 18 months after starting therapy\textsuperscript{29}. Failure in achieving or maintaining the goal of HbA1c level <7% with maximally tolerated of initial monotherapy by MET may be consider as a reason for necessity of combination therapy over time or prescribing more effective dose\textsuperscript{23}.

Previous studies have also found dosage up of MET as effective as adding another T2D medication with the probability of not achieving glycemic control and rate of glycemic control within 6 months. MET dosage up titration could be a preferable initial intensification strategy in patients there is a concern for gastrointestinal adverse effects, in which case adding a T2D medication might be preferable. Our results showed monotherapy by MET or additional T2D medication post initial MET such as TZD or SUL reduced HbA1c level during long follow up. HbA1c levels decreased significantly in the group of combination therapy of MET and SUL. Achievement of the lower level of HbA1c was higher in combination of MET with SUL than combination of MET with TZD or MET alone. There is some evidence for initial combination therapy due to the greater initial reduction of HbA1c than can be provided by MET alone\textsuperscript{30}.

T2D patients do often show surprisingly strong reductions in HbA1c with MET based combination oral treatment approaches. A recent report showed that even with base-line HbA1c >11%, the combination of MET with a SU, or TZD was associated with reduction in HbA1c from 11.6% to 6.0%\textsuperscript{31}. A 32-week study of the combination of TZD with MET in patients with mean baseline HbA1c 8.9% showed a mean HbA1c reduction of 2.3%, and an open-label cohort with baseline HbA1c 11.8% had a reduction in HbA1c to 7.8%\textsuperscript{32}. TZD, improves glycemic control primarily by increasing peripheral INS sensitivity in patients with T2D, whereas MET, exerts its effect primarily by decreasing hepatic glucose output.
SU have been the first oral antidiabetic medication to be introduced clinically for treatment of T2D\textsuperscript{33}. According for around 25% newly diagnosed T2D patients have used from this medication either as a first-line recommended choice or in combination with another glucose-lowering medication\textsuperscript{34}. This medication has widely prescribed as the second-line after MET in the UK\textsuperscript{35}. Result from our study showed prescribing this medication as the second line after INS for 32.5% of patients during 25 years from 1985-2010. In our study HbA1c levels in presence of SU increased by increasing SU up to 0.045 units (...), decreased 0.17 units by combination of SU and TZD (P = 0.001) and decreased 0.093 units by combination of MET and SU (P < 0.001). We have demonstrated in this study that prescribing SU plus INS will not reduce HbA1c significantly.

The result of association between the effects of OAD medication on HbA1c level from sixty-two trials showed the significant reduction in HbA1c for all agents from this family (SU), ranging from 0.43% to 1.29% with the highest range for glibenclamide\textsuperscript{5}. Result of another study showed the range from 24% to 88% for achieving HbA1c < 7% in the first year after therapy for the highest to lowest baseline HbA1c in patients whom used initial SU. Most OAD agents lowered HbA1c levels by 0.5 - 1.25% within the first 4 to 6 months, whereas SU lowered HbA1c levels by 1.25%\textsuperscript{21}. Result from nine trials of SU monotherapy\textsuperscript{36} suggest this medication effect for reducing HbA1c by around 1.51% than placebo and 1.5% in combination with MET\textsuperscript{37}. Result from a systematic review with including six trials of SU reported that this treatment reduced HbA1c by around 1.5% as monotherapy with different doses and in combination with other oral medications reduced HbA1c by 1.6% compared with placebo\textsuperscript{38, 39}. The greatest effect of combination with INS was seen for glibenclamide with reduction rate around 0.6% than INS treatment alone but resulted in lower using dose\textsuperscript{40}. Result from recently study showed adding SU to OAD (four trial) lowered HbA1c by 1.62% compared with the other treatment and adding to INS (17 trials) lowered HbA1c by 0.46% with lower dose of INS and higher dose of this medication didn't reduce HbA1c more than lower doses\textsuperscript{33}. Previous studies confirmed that initial monotherapy by SU often fails to achieve and maintain HbA1c goals of < 6.5% and < 7.0%\textsuperscript{41}. After 1 year therapy by this medication only 33% of patients attained an HbA1c < 6.5% and 54% attaining an HbA1c < 7.0%\textsuperscript{22}.

Hyperglycemia in T2D patients is result of pathogenic defect including insulin resistance and pancreatic islet cell dysfunction\textsuperscript{42}. Progressing of the disease appears mainly to the result of a continuing loss of B-cell function. Treatment with diet, insulin, sulfonylurea, or metformin is known to improve glycemic control\textsuperscript{43}. When oral anti diabetic drugs cannot reduce the level of HBA1c to target level, most patients will need INS\textsuperscript{12}. There is a group of patients with T2D who do not respond sufficiently to the currently available glucose-lowering drugs which are prescribed, despite higher levels of treatment regimens including MET and/or SU.
An interesting potential combination is that of an INS with a TZD.

INS therapy is considered as the most effective treatment with the strongest association for treatment of persistent poor glycemic control. The observed result from our study indicated that individuals with history of using INS could better manage their treatment because these patients had a significantly lower change in mean HbA1c level (estimate of -1.049) when compared to patients with history of using OADs regimen. Single active agent’s effect for reduction of HbA1c was significant for INS superior to 40% of other antidiabetic medications. Retrospective and prospective observational studies reported final HbA1C values from 7.9 to 8.2% after starting INS for patients with baseline level of HbA1C ranged between 8.8 and 10.2%. Patients with basal HbA1c >8.8% especially under treatment with more than two OADs, have small chance for achieving good glycemic control with adding INS to their regimen. Early initiation therapy regimen with an intensive INS-based should be prescribed for these patients. Two systematic reviews that evaluated the effect of INS have indicated that reduction of HbA1c to target level (HbA1c<7%) is achieved in 35–54% of T2D patients. Other associated factors as age, body mass index, HbA1c at baseline and time of adding INS to OADs have been reported to influence on treated of T2D patients with INS. Chance of achieving glycemic target will decrease or becomes more difficult with delaying usage of INS. One meta-analysis indicated to greater reduction in HbA1c level in patient with earlier stage of treatment only with INS or adding that to one OAD medication with a lower risk of hypoglycemia than for patients who are treated with two OADs. Treatment by TZD alone was not significant in decreasing HbA1c level over time in our study. It can be indicated that there was no significant differences in changes of HbA1c values for the different levels of this variable over the time. TZD plus INS therapeutic regime led to a significantly (0.0009) less HbA1c levels. Treatment with combination of INS and TZD was predicted to have more HbA1c reduction followed by SU plus TZD (0.0014) and MET plus TZD (0.0382). The results of our study are consistent with the results of previous study that indicated significant effect of this medication for lowering A1C levels by 1.0% within the first 4 to 6 months after treatment. Age has been reported to influence glycemic control in T2D being treated with INS.

INS treatments and TZD plus INS therapeutic regiments led to a significantly less HbA1c levels in our study. In a study comparing the addition TZD with the addition of INS in 101 people receiving SU and MET with baseline HbA1c >10%, HbA1c fell from >11% by >4% compared with <4%, respectively, and the INS plus TZD treatment was associated with less weight gain and hypoglycemia. Estimated result from the mixed effects model showed the significant effects of treatment by INS and its combination with TZD on mean HbA1c level. Evidence from the prospective and clinical diabetes studies emphasize on benefit of clinical effect.
of early intensive treatment by INS on cell function and reducing glycemic level\textsuperscript{52}.

Some of the studies found that older individual with longer diabetes duration had better glycemic control\textsuperscript{53}. This finding in our study and somewhat is in contrast to the result of other prospective diabetes study which suggested that age is not significant main effect in predicting glucose control or represented the significant effect of younger age besides other factors as higher base line level of HbA1c and longer diabetes duration over time for delayed response to therapy without achieving glycemic control and high level of T2D complications especially retinopathy\textsuperscript{53, 54, 55}. However HbA1c over time has been indicated with different pattern between age groups in a study but another study indicate no significant difference for glycemic level between age groups\textsuperscript{53, 54}. We couldn’t confirm a significant association between the level of HbA1c and sex but other previous studies represented poor glycemic control in female gender\textsuperscript{55}.

**Strength and limitation**

A major strength of this study is that it was conducted in patients with T2D from a routine primary care setting in the Netherlands, and the results are therefore directly relevant for daily clinical practice. A wide range of clinical data was included to characterize the different effect of antidiabetic medication on HbA1c level. Other studies, mostly been cross-sectional with small sample size and short follow up, by using self-reported medication that may cause some misclassification in measured treatment regimens and have examined the relationships between T2D and one or more factors.

HbA1c is the most important endpoint for evaluating response to medication, and we were able to examine long-term endpoints in our studies. Previous studies suggested more research over time and repeated measurements that can be more informative for more conclusions with respect to the findings of other cross-sectional studies. So, using frequently monitoring of HbA1c and adjustments of glucose-lowering drugs and using the various variables together in one model and taking the relationships between them into account, the large sample size, and follow-up duration are other strengths of this study.

Considering the limitation and suggestions of other previous studies of, this study is one of the first current based on longitudinal approach and mixed models that enable the researcher to compare the groups and find factors associated with changes in HbA1c by repeated measurement HbA1c.

However, there are also some limitations to be considered. Because this study analyzed a real life primary-care cohort of patients, the follow-up period was long. A longer follow-up period would lead to an increased risk of selection bias due to losses during the follow-up. The number of patients included in the study de-
creased during follow-up. This is partly because there are no data covering the follow-up for patients. In addition, some patients may have left the cohort because they died, moved out of the region, were referred to secondary diabetes care, or moved into a care home for the elderly.

**CONCLUSION**

The results of our study are in agreement with several previous studies show that initial monotherapy with either MET or TZD alone or even a combination of INS with MET, or INS with SU fails to achieve a low HbA1c level in long term. INS remains an important part of treatment for T2D, either alone, or in combination with TZD, and is certainly a needed option for many patients. To prohibit development of INS resistance, a combination of TZD with MET or SU seems as the most effective alternative in reducing HbA1c. Our result suggests that not every patient may benefit from the same type of glucose management, and there may be certain patients for whom different care strategies and more stringent HbA1c targets are appropriate.
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