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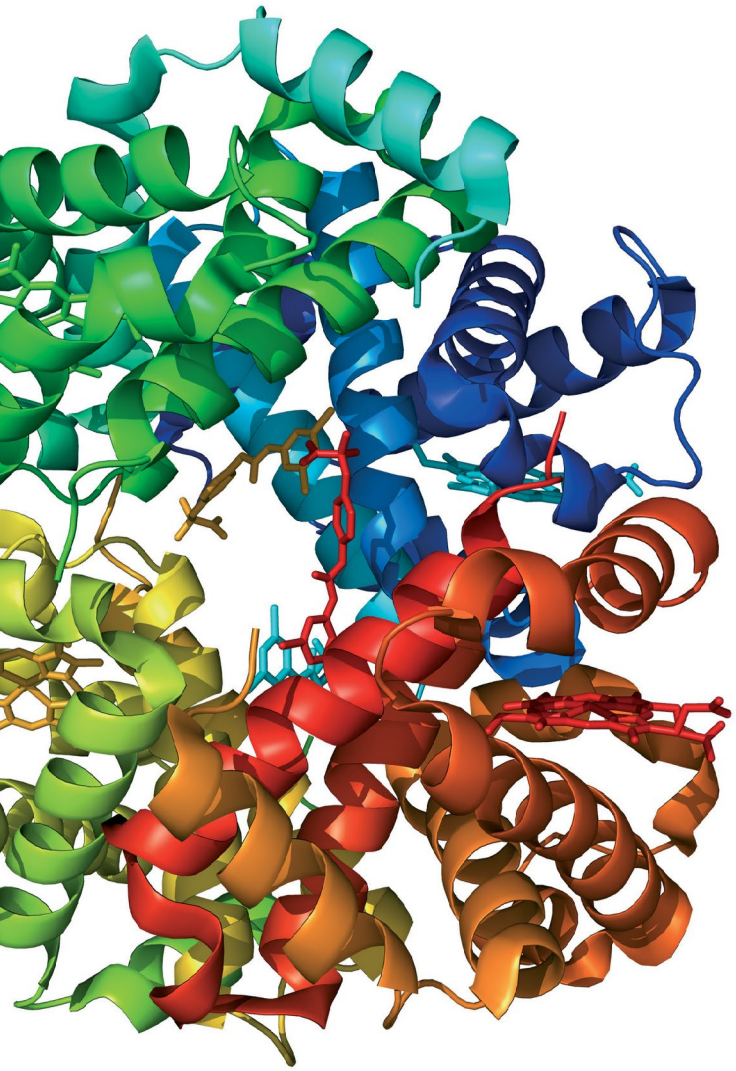

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Contents

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Diabetes mellitus and COVID-19: factors associated with bad prognosis

ABSTRACT

Diabetes is a risk factor for bad prognosis of COVID-19. Many mechanisms can explain the bad prognosis of COVID-19 in diabetics, but they remain hypothetical. The high prevalence of diabetes on the African continent, particularly in North Africa (12.2%), constitutes a threat of increased morbidity and mortality linked to COVID-19. We must pay particular attention to this fragile population, with more time and resources, especially for the elderly, obese or those with chronic complications of diabetes who have a high risk of developing severe forms. (Clin Diabetol 2020; 9; 4: 205–207)

Key words: COVID-19, coronavirus, diabetes

New pneumonia caused by a new coronavirus named SARS-CoV-2 appeared in China at the end of 2019 and quickly spread around the world (it approaches 5 million cases in 188 countries at the time of writing of this letter). The case fatality rate varied so much internationally, In China it was between 1 and 2%, in Europe it was around 9% in some countries and in Africa it is 3.2%. This disparity in rates is explained by the quality of screening and the nature of the population. We have noticed in Morocco, that the lethality rate went down from 7% in April to 2.7% in May, this decrease was proportional to the increase in the number of screening tests per day. In the literature, the most distinctive comorbidities related to a poor prognosis of COVID-19 pneumonia are cardiovascular (hypertension and ischemic heart disease), metabolic (diabetes, obesity), chronic or respiratory renal diseases [1]. Table 1 summarizes the main comorbidities associated with COVID-19 infection in Italy [2].

Diabetes is one of the most common comorbidities in infected patients, the second in China and Italy following hypertension [2–4]. Data from China have reported an increased incidence of COVID-19 in diabetics [1, 2]. However, a recent meta-analysis of 12 studies including a total of 2,108 Chinese patients concluded that diabetes paradoxically does not increase the risk of infection by the virus [5]. Regarding the impact of diabetes on the prognosis of the disease, only 6 of 12 Chinese studies have been able to provide information, and they confirmed that diabetes should be considered as a risk factor for a rapid progression and bad prognosis of COVID-19 [5]. Similarly, based on data from the Chinese center for disease control including more than 44,000 confirmed cases, the fatality rate was multiplied by 3 in the presence of diabetes (2.3% *vs.* 7.3%) [5]. In virology, this finding is not new, it has already been reported for other respiratory viral infections including seasonal flu, influenza A (H1N1) in 2009 and the two previous COVID infections: SARS in 2002 and MERS in 2012 [6, 7]. A recent French observational study was the first to focus on the predictors of severe forms in hospitalized diabetic patients; they were similar to those found in the non-diabetic population, such as age and obesity, but adding that the presence of complications of diabetes was positively associated with death. Insulin (the agent of choice to control glycaemia in hospitalized patients with COVID-19), like antihypertensive medications that interact with the renin-angiotensin-aldosterone system, is not a risk factor for severe form of COVID-19 [8].

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Many mechanisms can explain the bad prognosis of COVID-19 in diabetics, but they remain hypothetical, several factors have been mentioned in the literature:

— type 2 diabetes, often associated with obesity predisposes to a pro-inflammatory state via increased inflammatory mediators interleukin 6 (IL-6) and tumor necrosis factor α (TNF-α), which might facilitate the cytokine storms, which in turn appears to be the cause of the severe cases of COVID-19 pneumonias and of death in many patients;

— high mortality rate in the presence of chronic complications of diabetes, particularly macrovascular ones, was raised in Chines series and confirmed in the French study [8, 9];

— inflammation increases insulin resistance and the need for insulin, which affects glucoregulation and increases the risk of ketoacidosis. On the other hand SARS-CoV-2 could directly damage beta cells, this hypothesis has been raised to explain the large number of new cases of diabetes and also the need for high doses of insulin in infected patients. Indeed pancreatic beta cells express also the ACE2 receptor (the coronavirus binding site), such as lung, heart and kidney cells [3]. This hypothesis suggests screening for diabetes in patients with COVID-19 infection;

— hypercoagulability appears also to be the cause of the severe cases of COVID-19 pneumonias. It is known that diabetes is associated with hypercoagulability and endothelial dysfunction which is only partially corrected by optimal glycemic control. In addition the association with a procoagulant infection by the cytokine storms will increase hypercoagulability, this explains why the level of D-dimer was significantly higher in diabetic patients infected with the coronavirus in several series [9, 10]. Conversely, hypoglycemia increases also the inflammatory state and platelet reactivity, this is why strict glycemic control during coronavirus infection could be deleterious. So rigorous monitoring of capillary blood glucose and therapeutic adaptations are strongly recommended, especially in the case of treatment with hydroxychloroquine. The latter, with an unknown hypoglycemic action, is approved by most African medical societies for the treatment of SARS-CoV-2 in combination with azithromycin;

— finally, an even more worrying aspect of the interaction of two pandemics, is the asymptomatic character in the initial phase of the infection which was noted in the Chinese's series, with less fever, chills and shortness of breath. This could be responsible for a delay in treatment and an increase in the incidence of severe forms [9].

Table 1. Main comorbidities associated with COVID-19 infection in Italy [2]

Type of comorbidity (%)	
Hypertension	73.8
Diabetes mellitus	33.9
Ischemic heart disease	30.1
Atrial fibrillation	22.0
Chronic renal failure	20.2

Conclusion

In the light of these findings and the high prevalence of diabetes on the African continent, particularly in North Africa (12.2%), there is a threat of increased incidence of morbidity and mortality linked to COVID-19. In addition to chronic complications, the pneumonia caused by COVID-19 worsens the prognosis of our diabetic patients. We must pay particular attention to this fragile population, with more time and resources, especially for the elderly, obese or those with chronic complications of diabetes who have a high risk of developing severe forms. Without forgetting that COVID-19 infection is an opportunity to screen for diabetes. The future research will explain the behavior of COVID-19 in patients with diabetes.

Conflict of interests

The authors declare to have no conflict of interests.

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COVID-19 specialized diabetes clinic model for excellence in diabetes care: scientific perspective

ABSTRACT

While diabetes centers are well established by the Ministry of Health, there is no separate specialized diabetes clinics for COVID-19 patients (SDCs). There are several clinical diabetes centers throughout the Kingdom of Saudi Arabia, several of which have been developed through philanthropy funding; nevertheless, it is not obvious what distinguishes SDCs from a therapeutic viewpoint and what the potential would be for such centers. Through this context, we suggest a structure to direct the progress of SDCs. Defining protocols for wider adoption of SDCs as a means to enhance public safety and COVID-19 patient care efficiency (including consistency and satisfaction) and minimize health care expenses becomes increasingly essential when moving towards value-based sales and reimbursements away from service charges. It is wise to introduce innovative financial mechanisms to pay for diabetes that cannot be covered by fiscally limited private and university medical centers. We foresee potential clinical SDCs to be made up of a well-defined framework and six areas or foundations that act as basic guiding principles for the advancement of diabetes treatment skills that can be easily illustrated by stakeholders, including insurance facilities, consumers, payers and government departments. (Clin Diabetol 2020; 9; 4: 208–211)

Key words: COVID-19, specialized diabetes clinic, model for excellence, diabetes care

Introduction

Diabetes is a significant public health problem. Nearly 30 million individuals in the United States are diabetics [1]. Diabetes accounts for 12% of all deaths in the United States. Significant risks of diabetes lead to death and morbidity, with immense corresponding economic pressures [2]. The cumulative tangible and intangible annual cost of health care for this disease is estimated at 245 billion dollars [3]. Due to the economic and health challenges associated with diabetes,

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it is important to increase access to treatment and the way in which it is provided.

Centers of diabetes are well established by the Ministry of Health, but there is no specific description of the professional specialized diabetes clinics (SDCs). There are several clinical diabetes centers throughout Saudi Arabia, several of which have been developed through governmental funding; nevertheless, it is not obvious what distinguishes SDCs from a therapeutic viewpoint and what the future would be for such centers.

Thereunder, we suggest a system to direct the progress of clinical SDCs. With the move towards value-based payments and discounts, and away from utility charges, identifying protocols for wider adoption of SDCs as a means of enhancing public safety, increasing COVID-19 patient care efficiency (including consistency and contentment) and lowering health care costs are becoming crucial. It's wise to introduce innovative funding mechanisms to pay for diabetes that cannot be covered by financially limited private and university medical centers.

Three separate dimensions lead to success in diabetes treatment: (1) the delivery of clinical services through trained multidisciplinary medical teams headed through skilled diabetic professionals experienced with treating challenging, high-risk COVID-19 patients; (2) scientific performance that includes training, clinical study and student activity; (3) COVID-19 patient awareness, involvement and satisfaction with comprehensive quality care models.

The framework concept and perspective outlined in this article describes the fundamental components of developed SDCs that include exposure to the continuum of "state-of-the-art" of diabetes treatment.

Specialized diabetes clinics vision statement

A systematic SDC model will consist of an architecture and six realms or principles that act as basic guiding standards for building knowledge in diabetes treatment that can be readily communicated to stakeholders, including health care professionals, consumers, payers, and government departments.

Infrastructure

First, the concept of SDC requires the development of a sufficient infrastructure to function as a 'center.' Adequate personnel is a core component of this infrastructure to provide information on diabetes self-management to new and current COVID-19 patients; provide advice on the administration of injectable drugs; invest in diabetes--related technology (e.g., continuous glucose monitoring, insulin pumps, meters). Access and evaluation of glycemic data at the point of treatment during visits to facilitate appropriate improvements in therapy as indicated; explore nutrition, exercise, and usage of medications in relation to COVID-19 patient lifestyles; answer management-related phone calls, messages and e-mails, in a timely fashion. Delivering appropriate clinical behavioral wellbeing and social care, including recognition and referral to community resources; providing medical instruction to clinicians at the center and throughout the community, advising family members and other professionals, and resolving drug pre-authorization liability problems through third party policy. In the absence of such infrastructure a real SDC cannot work.

Principles for specialized diabetes clinics

Non-exclusive emphasis on high-risk people and open-door policy

With the current diabetes crisis SDCs must face the number of COVID-19 patients qualifying for treatment that frequently surpass capability owing to a nationwide scarcity of endocrinologists [4]. There are, 6500 endocrinologists in the United States, half of whom do not treat diabetes patients with COVID-19. Recent estimates reflect the actual scarcity of 1,500 clinically engaged adult endocrinologists, a number that is predicted to increase [5].

This problem can be addressed by two potential, preferably complementary pathways.

Systematic training of nurses trained with diabetic treatment will help improve coverage for those who may profit more from accessing treatment in the SDCs. This covers all people with type 1 diabetes and those with inadequate glycemic regulation, compromised hypoglycemia, various complications, cystic fibrosis, post-transplant diabetes, atypical types of diabetes, and complicated medical dilemmas. This method helps the emphasis to be on population control and risk stratification prior to referral.

At the same moment, whenever practicable, an "open door" strategy will make sure that COVID-19 patients require treatment were not rejected. For this reason specialized professionals such as nurses, medical assistants, and allied health practitioners (clinical pharmacists, registered nutritionists, and accredited diabetes educators) may be qualified and equipped to play a greater coordinating function in the control of COVID-19 patients receiving treatment at the SDC.

Collaboration through the medical system to direct treatment

Structured and consistent contact and continuity of COVID-19 patient care is another critical factor in delivering efficient and quality care of COVID-19 patients across the continuum of diseases identified in SDC. Co-

ordinating treatment through patient-centered medical homes (PCMHs) starts with treatment arrangements that define requirements, duties and commitments at the point of admission to primary treatment [6].

Pre-consultation exchanges. Pre-consultation exchanges resolve clinical issues that may not require in-person visits, such as the need for slight drug modifications. Addressing these issues in a timely manner compensates for the requirement for comprehensive in-person visits to the SDC and enables ongoing treatment by primary care physicians. It also reduces the waiting period for new COVID-19 patient consultations for those listed as having the greatest need. Reimbursement for pre-consult transactions focused on a perfected model offers a financial reward for the time invested in such consultations. In places where accredited diabetes educators and expert doctors are not eligible, online consultations (e-consults) can be used to assess the necessity for in-person visits [7, 8]. Many health care organizations have tested e-consultations or telehealth as emerging forms of treatment management [9, 10]. While e-consults are used more widely in pharmacy, they play a role in the treatment of diabetes.

It poses challenges. For instance, despite having experience of when and how to start insulin (or other intravenous) therapy, there is a shortage of resources for many policies and procedures (PCPs) to implement. Among many cases, e-consultations do not resolve the problem of limited supply of personnel to handle pre-authorizations and appeals against certain current diabetes insurance programs. E-consultations could also be a great opportunity to incorporate advice from experts with continuing education. The SDC could help to overcome these challenges.

Structured appointments and communal management

Some COVID-19 patients require a number of in-person visits to the SDC to initiate injection therapy, such as the use of various options or co-formulations to promote basal-bolus insulin or insulin pump treatment, or the use of professional or personal continuous glucose monitoring. Such COVID-19 patients will also go back to their PCP for a referral. Integrated heathcare and comprehensive patient management should be considered, once the problems of PCPs have been resolved. Continuity of treatment is maintained by the COVID-19 patient remaining in the PCMH with his or her PCP for day-to-day control while retaining access to the SDC.

Specialized diabetes clinics management may include communal management of diabetes. An example could be a COVID-19 patient who begins insulin at the SDC, returns to the PCP, and visits the SDC every 6 to 12 months.

Creative approaches

The use of technical innovations, including more articulate and unbiased online communications between suppliers of medical information, will render hospital care more effective around the board and spectrum of care, along with transition periods, such as from medical facility to home [11].

Proper treatment

The Unified Referral Network guarantees diabetes treatment is given effectively and securely by the SDC. As the bulk of cases should be assigned to SDC, free and continuing contact via their corresponding family physician ensures that both companies have the correct reviews and accessible participation in other areas of diabetes treatment. Two of them, the theoretically successful approach is to schedule monthly group research. Sessions for endocrinologists and partners providers where COVID-19 patients are presenting limited or more difficult performance problems should be addressed. Usage of other specialties linked to diabetes necessary for intensive diabetes treatment COVID-19 patients and suppliers in the establishment of screening or early diagnosis and treatment of diabetes-related complication. This can be achieved by positioning specialties that operate as a one-stop shop for COVID-19 patients. Convenience could be reached by scheduling on-line meetings with colleagues, the same day of screening and/or treatment of diabetic COVID-19 patients by multidisciplinary teams.

Healthcare information system

Constant emphasis improving consistency and engagement in the SDC is an important subject of clinical studies to improve pharmacology and the public medical treatments with diabetes mellitus. You should use the SDC, the Model of Health Care Education that Connects Persons, including details on the community in a consolidated including systematic manner database [12] or register that requires the evaluation to be carried out in interest. Such registries are optimally planned to fulfill the minimum criteria. Adherence to standards, for providing secure exposure for studies and many experts who will support the center in the extraction process will lead to significant clinical and epidemiological results. Determined COVID-19 patient details would allow feasible relations from SDC, science promotion and quality management approaches that can go beyond the United States to global groups.

The SDC will act as a sign of continuous improvement measures that aim to insure that the most relevant robust and up-to date treatment is given in the community requires modern technology and innovations of clinical research. Ideally, the Defence Centers of Excellence

will implement initiatives such as the iterative do-check method system for long term ventures [13] Kaizen Activities short-term development programs [14] and Lean Six Sigma (a process that depends on a joint effort by the team improving efficiency by consistently reducing waste reducing variation) [15]. When approaches are tested systematically, within the context of a health literacy network, successful approaches can spread quickly through the health sector then after that, by a group of SDC.

Determination of the outcome

A constant endeavor must be made to preserve the SDC preservation of good quality treatment through the development of verifiable evidence for analysis and feedback from other parties, such as funding associations or insurance providers, as well as COVID-19 patients. Present indicators, such as target-directed measures of HbA_{1c}, lipid and blood metrics. Pressure is necessary but of limited value. Scale, such as pause in development of complications severity of hypoglycemia, reduction in coronary condition, morbidity, death and general lifespan and the standard of life is going to be more critical than that measures by the researcher alone.

Awareness and distribution

Dissemination of results and professional practice SDC positively impacts the other organizations concerned about treatment of diabetes. Personal, regional, global conferences, grand rounds and small group meeting will do the utmost to encourage practices within and outside an agency. Grand rounds and small group meetings will do the utmost to encourage practices within and outside an agency. In Pennsylvania there is a Chronic Health Program, which funds and promotes training primary care procedures in the PCMH model and extension of the Public Healthcare Outcomes program, that supports the tele-mentoring of PCPs by diabetics, details of these partnerships [16, 17].

Quick "expert classes" that have information on the topic core factors in the treatment of diabetes can aid disseminating specialist treatment for diabetes outside isolation it's a SDC. Outside the structured educational courses, SDC had the chance to consult with an "initiate endocrinologist" programs under which the center's experts visit or create near ties with corresponding family physician at the same time or to other organizations to pass experience and grow it mutually advantageous ties.

Conclusions

Specialized diabetes clinics are an essential component in innovative approaches in cost-conscious health care distribution, interconnected, COVID-19 patient-centered treatment for people with diabetes. Optimizing health results when evaluating expense, efficacy and consistency can result in cost-effective treatment. The suggested SDC model for an effective and well thought-out solution. The network, combined with six main components, is a practical approach to the production of verifiable and transferable standards in treatment with large COVID-19 patient groups. The System may act as a prototype and a comparator for new and emerging versions. Emerging SDC in an effort to standardize and build good treatment for diabetes.

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C/T polymorphism of the rs7903146 nucleotide of the *TCF7L2* gene and the risk of developing diabetes mellitus type 2

ABSTRACT

Background. The continuously increasing incidence of type 2 diabetes mellitus (DMt2) is an important problem in current medicine. In addition to well-known environmental factors that may contribute to this disease, genetic factors may also play a role. One of the genes that may be responsible for an increased risk of DMt2 development is the transcription factor *TCF7L2* gene. This work was aimed to assess a correlation between the rs7903146 polymorphism in the *TCF7L2* gene and age at DMt2 diagnosis, presence of obesity, arterial hypertension, and time elapsed from DMt2 diagnosis to the start of insulin therapy.

Methods. An analysis of the studied polymorphism was performed in 282 patients diagnosed with DMt2. Patients were divided into groups depending on the age of the onset of DMt2: group A (n = 82) — DMt2 diagnosis below the age of 40 years, group B (n = 100) — DMt2 diagnosis between the ages of 40 and 60 years, group C (n = 100) — DMt2 diagnosis above the age of 60 years.

Results. In group C, there was a significantly lower number of patients with a TT genotype of the studied polymorphism compared to the combined group A + B ($P < 0.05$).

Conclusions. There is a correlation between the rs7903146 polymorphism in the *TCF7L2* gene and age at DMt2 diagnosis. No correlation has been demonstrated between the studied polymorphism and the presence of obesity, arterial hypertension, and time elapsed from DMt2 diagnosis to the start of insulin therapy. (Clin Diabetol 2020; 9; 4: 212–218)

Key words: diabetes type 2, *TCF7L2* gene, age, polymorphism

Introduction

The continuously increasing incidence of type 2 diabetes mellitus (DMt2), spanning all age groups, which is currently estimated at approximately 8%, classifies DMt2 as the most common social disease. The incidence of diabetes in the Polish population is comparable to the average values observed worldwide [1]. The most numerous population of patients with diabetes are people aged 40 to 59 years. In the pathogenesis of DMt2, in addition to known environmental factors, genetic factors which have been extensively studied for several years play a role. The genome-wide association study (GWAS) has identified more than 60 loci related to a different degree with the risk of DMt2 development, mainly due to the effect on pancreatic beta cell function, insulin resistance and obesity [2]. The most well-known gene that can contribute to DMt2 development is the transcription factor 7-like

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2 gene (TCF7L2). The association of polymorphism in this gene with DMt2 development has been demonstrated for the first time in an Icelandic population in 2006 [3]. In subsequent studies, this relationship was confirmed in various ethnic groups [4–9]. The strongest association with the risk of developing DMt2 has been demonstrated for the T allele of the rs7903146 polymorphism in this gene. The odds ratio of DMt2 development in carriers of a single T allele is about 1.4, while in people with TT genotype, the chance of DMt2 development is about twice as high as in people without an unfavorable allele [10].

The product of the TCF7L2 gene is the TCF7L2 transcription factor, an important element of the intracellular WNT signaling pathway which plays an important role in many basic physiological processes such as embryonic development, stem cell maintenance, cell proliferation and migration, and oncogenesis [11]. While unfavorable polymorphisms of the TCF7L2 gene in many studies have been associated with a higher risk of DMt2, a similar relationship has not been shown for type 1 diabetes, maturity onset diabetes of the young (MODY type diabetes), and persistent diabetes in newborns [12, 13]. Nevertheless, there was a significant correlation between the rs7903146 polymorphism in the *TCF7L2* gene and the risk of latent autoimmune diabetes in adults (LADA) [12, 14]. Other polymorphisms of this gene were associated with an increased risk of gestational diabetes [15, 16]. In many studies, reduced insulin secretion was demonstrated in carriers of the T allele of rs7903146 polymorphism in the *TCF7L2* gene both in diabetic patients and in healthy subjects, which was also associated with a higher risk of DMt2 development [17–19]. The unfavorable polymorphism in the *TCF7L2* gene leads to impaired conversion of proinsulin to insulin, which most probably results from the regulating effect of TCF7L2 on the expression of the genes for convertase 1 and 2, responsible for this conversion [20, 21]. The *TCF7L2* gene polymorphism may also affect carbohydrate metabolism by affecting the incretin system. It has been shown that the production process as well as the mechanism of action of glucagon-like peptide 1 (GLP-1) are largely regulated by the WNT signaling pathway [22]. Since the TCF7L2 transcription factor gene is expressed not only in pancreatic and intestinal cells, but also in the liver, brain, skeletal muscle, adipose tissue, and bones, the effect of this gene's polymorphism on carbohydrate metabolism may include various pathophysiological pathways leading to the development of DMt2.

The influence of genetic factors on the age of patients in which DMt2 is diagnosed is not well understood. The relation between TCF7L2 polymorphism and the presence of hypertension or obesity also remains inconclusive, which additionally increases the risk of adverse cardiovascular events. Since the time from the diagnosis of the disease to the implementation of insulin therapy varies between patients, an interesting issue seems to be the assessment of the impact of genetic factors on the above-mentioned period, which indirectly reflects the time from the onset of the disease to the failure of beta cells. Due to a number of doubts regarding the influence of genetic factors on the development and progression of DMt2, the aim of this study is to evaluate the association of the rs7903146 polymorphism in the *TCF7L2* gene with:

— the age at DMt2 diagnosis;
— co-existence of hypertension and obesity in patients with DMt2 diagnosed in various age groups;
— a period of insulin independence.

Methods

The study included 282 patients with DMt2. Patients were divided into three groups depending on the age of the patients in which DMt2 was diagnosed:

— group A (n = 82) patients with DMt2 diagnosed before or at the age of 40 years;
— group B (n = 100) patients with DMt2 diagnosed between the ages of 40–60 years;
— group C (n = 100) patients with DMt2 diagnosed at or after the age of 60 years.

The inclusion criterion was:

— age ≥ 35 years, and
— effective treatment with diet or oral drugs/insulin ≥ 6 months.

The exclusion criterion was difficult logical contact with the patient.

After receiving written informed consent to participate in the study, a 4.9 ml blood sample was collected from each patient from the ulnar vein into S-Monovette test tubes (Sarstedt) containing 6.4 mg of potassium edetate. After 30 minutes from the time of collecting the biological material, the test tubes with blood were centrifuged (3,000 rpm, 10 min) and then stored at –20°C until DNA isolation, no longer than 6 months. Polymerase chain reaction (PCR) and allele identification were performed on a 7300 Real Time PCR System (Applied Biosystems). Genotyping of the C/T polymorphism of single nucleotide rs7903146 of the TCF7L2 gene was performed with fluorescence-labeled probes using TaqManPre-designed SNP Genotyping Assay (Applied Biosystems).

All results obtained were subjected to statistical analysis using the software package Statistica 9.0. Continuous parameters were expressed as means with standard deviation (SD), and categorical variables were presented as numbers and percentages. Normality of

Table 1. General characteristics of the studied population

	Group A (n = 82)	Group B (n = 100)	Group C (n = 100)
Age, years (SD)[1]	54.2 (11.5)	62.5 (8.6)	73.8 (7.3)
Gender M/F[2]	48/34	60/40	38/62
Age at diagnosis of DMt2 (SD)[1]	36.9 (4.5)	51.0 (4.7)	67.5 (6.1)
Duration of DMt2, years (SD)[1]	17.4 (11.0)	11.6 (8.0)	6.3 (5.6)
BMI [kg/m^2] (SD)[2]	31.5 (5.2)	30.7 (5.0)	29.0 (4.4)
Waist circumference [cm] (SD)[3]	109.0 (12.0)	106.5 (12.3)	104.3 (12.4)
Arterial hypertension [n (%)]	59 (72%)	75 (75%)	74 (74%)
Dyslipidemia [n (%)]	39 (48%)	61 (61%)	56 (56%)

SD — standard deviation; M/F — male/female; DMt2 — type 2 diabetes mellitus; BMI — body mass index
[1]Between each group, P < 0.05; [2]group C *vs.* group A and group B, P < 0.05; [3]group A *vs.* group C, P < 0.05

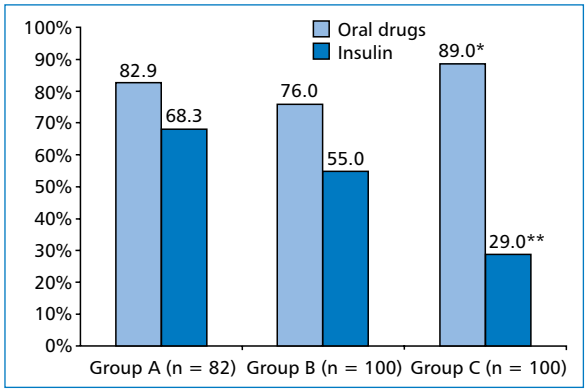

Figure 1. Method of therapy in individual study groups. *Group C *vs.* group B, P = 0.026; **group C *vs.* group B and group A, P < 0.001

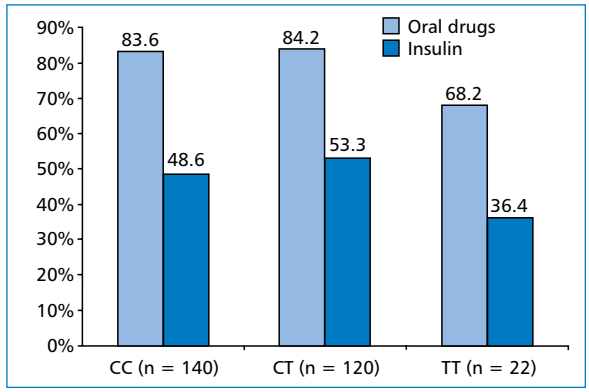

Figure 2. Method of therapy in the carriers of particular genotypes in the entire study population. For all P = NS

data distribution was tested with Shapiro-Wilk test. Comparative analysis between groups was performed using the t-Student test for continuous variables and the χ^2 for dichotomous parameters. P values < 0.05 were considered statistically significant.

The study design received a positive opinion from the Bioethical Committee of the Medical University of Silesia in Katowice.

Results

The general characteristics of the studied population divided into three analyzed groups are presented in Table 1.

Figure 1 shows the mode of therapy in individual groups divided into oral drugs and/or insulin. Statistical analysis showed that in group C, insulin therapy was statistically less frequently used than in other groups (P < 0.001) and oral therapy was more often used than in group B (P = 0.026).

A similar analysis regarding the method of therapy was carried out in relation to patients who are carriers of particular genotypes. In this analysis, there was no statistically significant difference in the mode of therapy between the CC, CT, and TT genotypes (Fig. 2). Similar results were obtained in individual groups A, B, and C.

Table 2. Frequency of particular genotypes in each study group

Group	CC	CT	TT
Group A (n = 82)	37 (45.1%)	35 (42.7%)	10 (12.2%)
Group B (n = 100)	45 (45%)	46 (46%)	9 (9%)
Group C (n = 100)	58 (58%)	39 (39%)	3 (3%)

Table 2 presents the frequency of individual genotypes in the studied groups. The rarest genotype in all studied groups was the TT genotype.

An additional analysis was made for the A + B and B + C combined groups and the frequency of occurrence of particular genotypes was compared to the remaining group. There was no statistical difference in the frequency of particular genotypes between the B + C

Table 3. Frequency of particular genotypes (group A *vs.* group B + C)

Group	CC	CT	TT
Group A (n = 82)	37 (45.1%)	35 (42.7%)	10 (12.2%)
Group B + C (n = 200)	103 (51.5%)	85 (42.5%)	12 (6.0%)

Table 4. Frequency of particular genotypes (group A + B *vs.* group C)

Group	CC	CT	TT
Group A + B (n = 182)	82 (45.1%)	81 (44.5%)	19 (10.4%)
Group C (n = 100)	58 (58.0%)[1]	39 (39.0%)	3 (3.0%)[2]

[1]*vs.* group A + B, P = 0.051; [2]*vs.* group A + B, P = 0.047

combined group and the A group (Table 3). Table 4 presents the frequency of individual genotypes in the A + B combined group compared to the C group. In the C group, a significantly lower percentage of subjects with the TT genotype was found compared to the A + B combined group (3.0% *vs.* 10.4%, P = 0.047). There was also a tendency for the CC genotype to be more frequent in group C compared to the combined group A + B (58.0% *vs.* 45.1%, P = 0.051).

In the analysis of the percentage of patients with the CC genotype compared to patients who were carriers of at least one T allele, no statistically significant differences were found between the studied groups.

In a similar analysis comparing the A + B combined group and the C group, a trend towards a less frequent genotype with at least one T allele in the C group than in the A + B combined group was demonstrated (42.0% *vs.* 54.9%, P = 0.051). There was no significant difference in the frequency of individual genotypes between the B + C combined group and the A group.

In the studied groups, there was no difference in the mean body mass index (BMI) among the carriers of individual genotypes. An additional analysis of the percentage of overweight and/or obese patients in individual groups did not show any relation to the studied polymorphism.

The CC and CT genotype carriers showed significantly lower BMI [kg/m^2] in group C compared to group A (for CC genotype 29.3 *vs.* 31.5, P < 0.05, for CT genotype 28.6 *vs.* 32.2, P < 0.05) as well as in group C compared to group B (for CC genotype 29.3 *vs.* 31.0, P = 0.05, for CT genotype 28.6 *vs.* 30.7, P = 0.06). This relationship was not found among carriers of the TT genotype.

The mean time from the diagnosis of DMt2 to the implementation of insulin therapy did not differ in the

Table 5. Mean time (SD) from diagnosis of DMt2 to implementation of insulin therapy (in years)

Group	CC	CT	TT
Group A (n = 82)	9.4 (6.8)	12.4 (10.8)	4.8 (8.2)
Group B (n = 100)	8.8 (6.9)	8.4 (8.0)	5.8 (5.7)
Group C (n = 100)	3.3 (4.0)[1]	2.2 (3.6)[1]	–

[1]Compared to group A and group B, P < 0.05

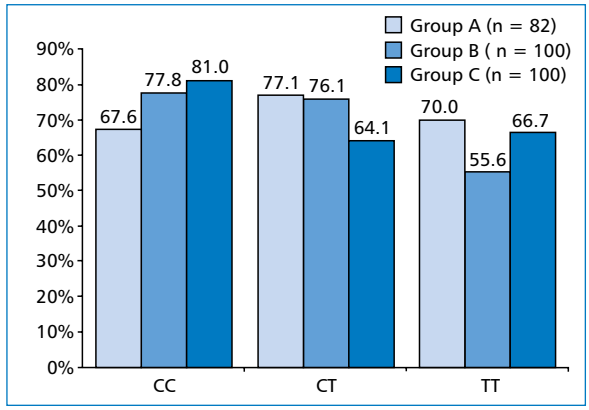

Figure 3. The co-occurrence of hypertension in the study groups

carriers of individual genotypes in each study group. In the carriers of the CC and CT genotype, there was a statistically significantly shorter time from the diagnosis of DMt2 to the implementation of insulin therapy in group C in comparison to both group A and group B (P < 0.05). A similar analysis could not be performed for carriers of the TT genotype due to the fact that there was only one patient in group C with this genotype in whom insulin therapy was used (Table 5).

There was no statistically significant relationship between individual genotypes and the co-occurrence of hypertension in the study groups (Fig. 3).

Discussion

In the presented study, the analysis of the gene polymorphism was performed in the groups of patients differing in the age of DMt2 diagnosis. According to the data of the International Diabetes Federation, the largest group of patients with diabetes are people aged 40 to 59 years. Patients with DMt2 diagnosed in this age range were included in group B, patients with disease diagnosed before this period were included in group A, and patients with disease diagnosed after this period were included in group C. There were 82 patients qualified to group A, 100 patients qualified

to group B, and 100 patients qualified to group C. The lower number of subjects in group A resulted from the lack of a sufficient number of patients diagnosed with DMt2 at or before the age of 40 years at study sites in the planned period, which is associated with the phenotype of DMt2, which in most cases affects people over 40 years of age.

The division of the studied population into the three groups defined above results in some differences in the general characteristics of the population studied. In the group of people diagnosed with DMt2 after the age of 59, a significantly smaller percentage of men was found than in other groups, which probably results from the longer average life expectancy of women in relation to men in the general population. The division of the population used in the study also influenced the average duration of diabetes, which was the longest in patients diagnosed earlier in life, and the shortest in the group of patients diagnosed after the age of 59.

The TCF7L2 transcription factor gene is one of the known candidate genes that may contribute to DMt2 development. The rs7903146 polymorphism of this gene results in the presence of three genotypes in the population: CC, CT, and TT. In all of the studied groups, the smallest percentage of patients were carriers of the TT genotype (from 3% in group C to 12.2% in group A). This result is similar to the data for the Caucasian population, for which the frequencies of particular genotypes are as follows: CC — 54.9%, CT — 34.5%, and TT — 10.6% [23].

The age of DMt2 onset is an important factor affecting the quality and life expectancy of patients. Because the duration of diabetes is directly related to the development of its complications, the diagnosis of the disease at a younger age has an adverse effect on the future fate of these patients. It was shown that patients with early diagnosed diabetes require the implementation of insulin therapy within three months of the diagnosis significantly more often than those diagnosed later in life. In this group of patients, the presence of microalbuminuria was also more frequent [24]. In another study, it was found that in the group of patients with the earlier diagnosis of the disease, the relative risk of macrovascular complications is twice as high as in the group of patients with the later diagnosis of diabetes [25].

Unfavorable genetic factors may be one of the reasons for earlier development of DMt2. In a study on a Mexican-American population, there was no statistical significance regarding the influence of the *TCF7L2* gene polymorphism on the age of onset of DMt2, although a trend towards such dependence was observed ($P = 0.055$). A similar result was obtained for the rs12255372 polymorphism of the *TCF7L2* gene, also considered one of the main genetic factors that may contribute to DMt2 development [26]. In another study on a Caucasian population, a relationship of the unfavorable T allele of the studied polymorphism with the earlier onset of DMt2 was found [27].

In the present study, it was shown that in the group of patients with DMt2 diagnosed before the age of 60 years, the presence of TT genotype is more frequent than in the group of patients with late diagnosis of DMt2. In turn, in the carriers of at least one T allele, a trend towards more frequent occurrence of this allele was demonstrated in the group of patients diagnosed with DMt2 before the age of 60 years. This may indicate an adverse effect of the T allele, and in particular the TT genotype, on the risk of developing diabetes earlier in life.

One of the most common comorbidities among patients with DMt2 is arterial hypertension. In the present study, in all age groups, the proportion of patients with hypertension was over 70% and did not differ significantly between the groups. There was no statistically significant relationship between the occurrence of hypertension and the studied polymorphism. Both DMt2 and arterial hypertension are known risk factors for macro- and microangiopathy, causing increased mortality among these patients. The ultrasound measurement of the intima–media thickness in the common carotid artery is considered to be a marker of subclinical atherosclerosis and can be used for identification of patients who are at increased risk of macroangiopathy, such as stroke or myocardial infarction. Bartman et al. showed that microvascular complications in patients with type 2 diabetes are independently associated with the carotid plaque score but not carotid intima–media thickness. So the study of carotid plaque score may serve to identify patients at risk of microvascular complications in patients with DMt2 [28].

The analysis of the method of therapy in particular groups in this study, including oral drugs and insulin therapy, showed that patients with diabetes diagnosed up to 60 years of age required the implementation of insulin therapy significantly more often than those patients diagnosed at an older age. This fact most probably results from a longer duration of the disease in this group of patients compared to patients diagnosed with DMt2 at older age. In our study, there were no relationships between particular genotypes and the method of therapy in the entire studied population and in individual groups. It was shown that the studied polymorphism had no effect on the mean time from DMt2 diagnosis to the implementation of insulin therapy in all the groups studied. It is worth noting that

in the group of patients diagnosed with DMt2 after the age of 59 years, the average time from diagnosis to implementation of insulin therapy was shorter than in the remaining groups, despite the shortest duration of the disease in this group. This may be due to a natural reduction in pancreatic beta cell function with age. Based on the results of this study, it can be concluded that the late age of diagnosis of DMt2 is associated with the need for earlier implementation of insulin therapy. However, due to the small number of patients treated with insulin in group C, further studies are needed to assess the possible effect of the studied polymorphism on DMt2 treatment.

Obesity is one of the main risk factors for the development of DMt2. A higher BMI in patients with DMt2 diagnosed before the age of 40 years may be one of the reasons for the earlier manifestation of diabetes in these people. On the other hand, in group C, a lower BMI can be considered as a factor protecting patients with a predisposition to developing DMt2 against earlier onset of the disease. The study did not show a relationship between the studied polymorphism and the occurrence of overweight and/or obesity in all the examined groups. The results of other studies evaluating the effect of the rs7903146 polymorphism of the *TCF7L2* gene on body weight are inconclusive [29, 30].

The presented study is an attempt to better understanding the role of *TCF7L2* polymorphism in pathogenesis of DMt2. There is no evidence that *TCF7L2* polymorphisms can serve as predictors of progression disease or influence response to pharmacotherapy. In spite of the ambiguous impact of this polymorphism on a number of clinical aspects of DMt2 patients, TCF7L2 seems to be one of the most important genetic factor in pathogenesis of DMt2. The impact of the polymorphism of *TCF7L2* may be affected by the origin of the study population. The major limitation of this study is the number of subjects enrolled. Because of low frequency of particular polymorphisms, studies comprising more patients are awaited.

Conclusions

The presented study showed that the proportion of patients with the TT genotype of the rs7903146 polymorphism of the *TCF7L2* gene is significantly higher among patients with DMt2 diagnosed before the age of 60 years than among those diagnosed at a later age. However, the relationship between the studied polymorphism and the BMI of the studied patients, the occurrence of overweight, obesity, coexistence of arterial hypertension, and the time from the diagnosis to implementation of insulin therapy has not been demonstrated. Nevertheless, the results should be treated with caution due to the relatively small number of patients. Further studies, both experimental and clinical, are necessary to determine the specific function of the *TCF7L2* gene in the regulation of carbohydrate metabolism, which may contribute to earlier identification of people who are at increased risk of developing DMt2 and improved treatment effects through appropriate therapy.

Conflict of interest

The authors declare to have no conflict of interest.

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The acute effect of a moderate intensity ergocycle exercise on the coagulation parameters in type 2 diabetes mellitus patients: a feasibility study

ABSTRACT

Background. Type 2 diabetes mellitus (T2DM) patients experience higher atherothrombotic risks that could lead to cardiovascular diseases, due to increases in coagulation activities. The role of exercise in altering coagulation activities among T2DM patients is still inconclusive.

This feasibility study aimed to evaluate the immediate effect of a moderate intensity ergocycle exercise, primary on the coagulation parameters and secondary on the systemic inflammation and blood glucose, in otherwise healthy T2DM patients.

Methods. Ten T2DM patients (64 ± 7 year, 40% female) performed a 30-minute moderate-intensity ergocycle exercise at 50–60% of heart rate reserved. Coagulation parameters (activated partial thromboplastin time [aPTT], prothrombin time [PT] and platelet count), erythrocyte sedimentation rate, and blood glucose were assessed before the exercise (T0), immediately after the exercise (T1), and 30 minutes post exercise (T2). One-way repeated measured ANOVA was used to assess the outcomes over time.

Results. All participants completed and adhered to the exercise protocol. There were increases in aPTT and PT but decreases in platelet count at T1 and T2 compared to at T0 ($P < 0.05$), indicating reduction in the coagulation activities. Those values, however, were still within normal ranges. The erythrocyte sedimentation rates were unaffected, while blood glucose decreased from 202 ± 35 mg/dL at T0 to 173 ± 33 mg/dL and 158 ± 30 mg/dL at T1 and T2 ($P < 0.01$).

Conclusion. The 30-minute moderate-intensity ergocycle decreases coagulation activities and blood glucose but does not affect erythrocyte sedimentation rates in T2DM patients. Future studies should focus on the chronic adaptation of the coagulation parameters after ergocycle training among a T2DM patients with coagulation impairments. (Clin Diabetol 2020; 9; 4: 219–225)

Key words: activated partial thromboplastin time, prothrombin time, platelet, erythrocyte sedimentation rate

Introduction

Type 2 diabetes mellitus (T2DM) patients are at significant risk of developing cardiovascular disease due to the impact of blood glucose elevation on the coagulation system, through a series of actions [1]. First, the increase of blood glucose could induce endothelial abnormalities and stimulate incomplete activation of the coagulation cascade [2]. Second, it causes glycation of proteins responsible for coagulation activities favouring hypercoagulation and stimulates pro-thrombotic states [3, 4]. Accumulatively, these events trigger the development of atherosclerotic lesions and occlusive thrombus which contribute to the pathogenesis of cardiovascular diseases such as ischemic heart disease and stroke [5]. Thus, hypercoagulation in T2DM patients should be monitored and controlled.

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Among the standard coagulation screening tests for monitoring hypercoagulation conditions in T2DM patients there are the activated partial thromboplastin time (aPTT), prothrombin time (PT) and platelet count performed [6]. The aPTT is used to assess the intrinsic and common coagulation pathway, while the PT assesses the coagulation pathway initiated by tissue factor, as well as the common pathway [7]. A prolonged aPTT and PT is a clinical indicator of fibrinolytic tendency, while a shortened aPTT and PT reflects hypercoagulation conditions [8]. Shortened aPTT and PT have been widely reported among T2DM patients [3, 9], along with increases in platelet activities [10]. These indicators, thus, are used for assessing and monitoring hypercoagulation among T2DM patients.

Exercise may play pivotal roles in altering coagulation activities [11, 12]. Evidence in the literature shows that exercise could increase both blood coagulation and fibrinolysis activities, which each causes opposite effects on the coagulation tendency [11]. The final effect of exercise may depend on the subject characteristics and exercise protocols [13]. In such, the effect of exercise may differ among different age groups or comorbidities and among different exercise intensities, types and duration. Specifically, transient hypercoagulation has been reported after an acute and strenuous exercise in individuals with coronary heart disease symptoms [12] and individuals with type 1 diabetes mellitus [11]. In contrast, increases in fibrinolytic activities were also reported among healthy individuals immediately after vigorous exercise [14], thus offer some protection against the risk of thrombosis and adverse cardiovascular events. These findings, however, may not be readily applied for T2DM patients, since these studies have been conducted in non-T2DM population and were based on vigorous-intensity exercise. Moreover, vigorous type of exercise is not recommended among T2DM patients and that the current physical activity recommendation for T2DM patients calls for a moderate intensity of exercise.

The heterogeneity of findings of the roles of exercise in altering coagulation activities in the literature, thus, substantiate the importance of studying the effect of moderate-intensity exercise on coagulation parameters. The ergocycle-based exercise was selected in this present study because stationary bicycling is one of the exercise types that is recommended for T2DM patients [16]. Thus, the primary aim of this study was to evaluate the feasibility and immediate effect of the moderate-intensity ergocycle exercise on the coagulation parameters, in a view to utilizing the exercise for improving hypercoagulation conditions in T2DM patients. The secondary aim was to study the immediate effects of the ergocycle exercise on systemic inflammation and blood glucose levels.

Methods

Participants

Ten otherwise healthy T2DM patients participated in this feasibility study. Patients were recruited from a public hospital in Yogyakarta Indonesia where they received regular care and diabetes medications from their physicians. They obtained clearance from their physicians to do regular exercise and participated in this study. At least three days prior to the commencement of the study, they were required to refrain from having performed any type of heavy physical activity.

Procedures and ethical clearance

The procedures used in this study were approved by the Ethics Committee of the Faculty of Medicine of Gadjah Mada University (KE/0565). Written informed consent was obtained from each participant. Participants were required to perform a warm-up exercise routine for ten minutes before the ergocycle exercise sessions. The warm-up exercise comprised flexibility exercises involving upper and lower extremities joints. The participants then were required to exercise on a bicycle ergometer to achieve free pedaling at workload up to two kilopounds (kp) until they established a regular and steady pedaling rate at 50 rpm. The workload was adjusted to reach the participants' heart rate at 50–60% of their heart rate reserved and was maintained at this level throughout the exercise period. The heart rate reserve was calculated by subtracting participants' resting heart rate from their maximum heart rate with a formula of 220 minus age. The exercise intensity was confirmed with the Borg Scale for ratings of perceived exertion. It was expected that participants would rate the scale at 10 to 12 during the exercise period to indicate the moderate intensity of exercise. The use of these methods has been validated and considered as a recommended approach for prescribing and monitoring exercise intensity in diabetic individuals [17].

The data collection was conducted on the exercise day with the support of two trained research assistants, two phlebotomists and one laboratory technician from a private clinical laboratory in Yogyakarta. The research assistants assessed participants' blood pressure, heart rate, and perceptions of exertion. The research assistants also ensured that participants adhered to the exercise protocol. Participants were required to take at least 10 hours overnight fast. The phlebotomists collected blood samples three times; (i) an hour after

a small standardized breakfast, prior to the ergocycle exercise, (ii) within 5 minutes after the ergocycle exercise completed, and (iii) 30 minutes after the exercise. The blood samples were taken with a clean vein puncture (20-gauge needle) from an antecubital vein under controlled venous stasis of 40 torr for less than 30 seconds. All vein punctures were taken in a reclined position.

Participants did not consume sugary drinks or food intake until the last blood sampling was performed. However, they could consume water or non-sugary drink, ad libitum, during the exercise and recovery period. Participants took their regular antidiabetic medication after the final blood sampling. Physicians were on site throughout the exercise and data collection period to monitor and ensure the safety aspect of the protocol, mainly for anticipating the signs of hypoglycemia that might occur.

Measures

Blood samples for the coagulation testing were prepared using citrate capillary with 3.2% sodium citrate and with a 1:10 dilution ratio to the specimen. Blood samples for other assessments were treated with EDTA. The complete methods for assessing the study outcomes and the corresponding normal reference range values are available in Table 1.

In addition to prothrombin time, for each participant, the international normalized ratio (INR) was calculated from the PT values according to standard practice. The data received from the laboratory technician were double-checked for ensuring the accuracy of the data. The range checks for data values were conducted and validated before the data were imported into a statistical program for analysis.

Analysis

The feasibility aspect was assessed by observing the ability and willingness of the participants in adhering to the exercise and data collection protocol. Repeated measured one-way ANOVA (RM-ANOVA) was carried out to assess the differences of outcome measures over time before (T0), immediately after (T1) and 30 minutes (T3) after ergocycle exercise. The Shapiro Wilk test and the sphericity assumption using the Mauchly's test were assessed. The Greenhouse-Geiser correction was used to interpret the within-subject effect for variables that did not meet the sphericity assumption. The pairwise comparisons using the Bonferroni post hoc test were explored if the within-subject effects were significant. Data were analysed using SPSS® v21.0 (SPSS Inc., Chicago, IL, US). A P-value < 0.05 represented statistical significance.

Table 1. Study outcomes, instruments and normal reference range

Study outcomes	Methods/ /Formula	Normal reference range
Coagulation parameters		
Activated partial thromboplastin time	Citrate plasma analysis with tromborel	23.0–30.2 second
Prothrombin time PT	Citrate plasma analysis with actin and $CaCl_2$	10.1–11.9 second
Platelets	Hydrodynamic focussing	150–440 thousand/uL
Systemic inflammation		
Erythrocytes sedimentation rate	Infrared sensor method with Westergreen value presentation	Male: 0–10 mm/hr Female: 0–20 mm/hr
Blood glucose	Spectrophotometry based on hexokinase method	< 110 mg/dL

PT — prothrombin time

Results

Participants characteristics

Participants' mean age was 64 ± 6.8, ranged from 47 to 74 years old. Forty percent of participants were female, Fifty percent had education level up to secondary education while the rest had tertiary education. Sixty percent were unemployed or retired and 80 percent were married. None was ex-smoker or smoker. Seventy percent had been diagnosed as having T2DM for more than 5 years. Ninety percent received oral medication and only one participant received insulin therapy. The mean of blood glucose prior to the exercise and an hour after the standardized breakfast was 210 ± 130 mg/dL. Most of them were also hypertensive with the mean of systole and diastole readings of 160 ± 20 and 85 ± 15 mm Hg, respectively. Lastly, most of them were considered overweight with the average body mass index of 26 ± 4 kg/m^2.

The feasibility of exercise and data collection protocol

All participants completed and adhered to the ergocycle exercise procedure and the three data collection. Participants were able to maintain the exercise intensity at moderate levels based on the prescribed intensity which was assessed with % heart rate reserve and rating of perceived exertion throughout the exercise period. No incident and sign of hypoglycemia were reported by participants and the physician. There was no missing data for all outcome measures at the three time-point.

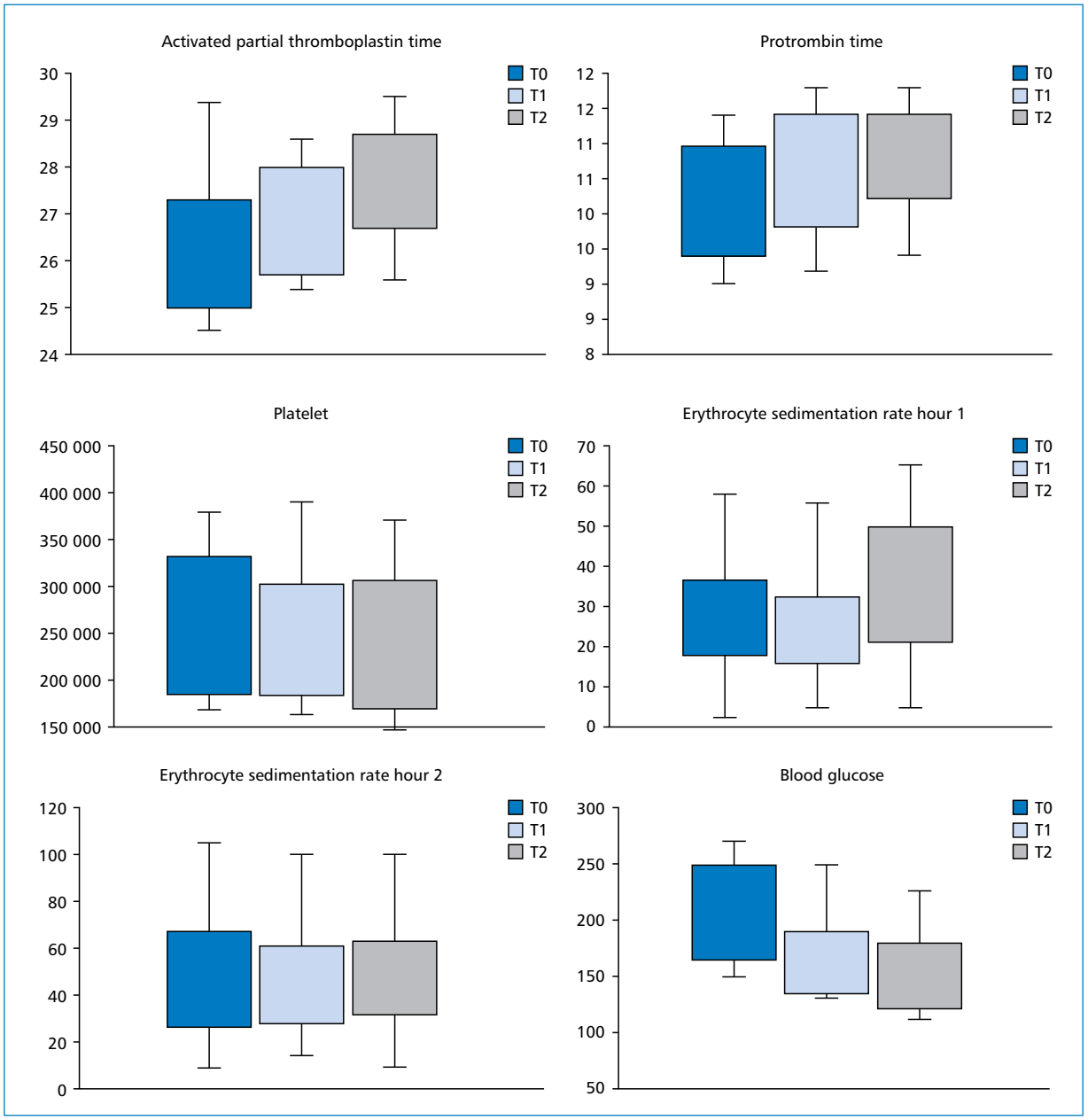

Figure 1. Ranges, interquartile, median and mean of the outcome measures before, immediately after and 30-minute post ergocycle exercise

Outcome measures at baseline, immediately after and 30 minutes after ergocycle exercise

Figure 1 illustrates the ranges, interquartile, median and mean, while Table 2 summarizes the means and standard errors of the outcome measures before, immediately after and 30-minute post ergocycle exercise, along with the results from the RM-ANOVA and post hoc analysis.

As shown in Table 2, all coagulant parameters increased immediately after the exercise. The values continued to increase during the recovery period except for platelet count which was the lowest at that period. Overall, these findings indicated a reduction in coagulation activity. The erythrocytes sedimentation rates were unchanged before and after exercise, while the blood glucose steadily decreased immediately and 30 minutes after exercise from the baseline value (< 0.01).

Discussion

This study assessed the feasibility of the application of a moderate intensity ergocycle exercise, among middle-aged and older T2DM patients to ensure the safety aspect of the exercise and study protocol. Our study demonstrated that the ergocycle exercise and

Table 2. The outcome measures before, immediately after and 30-minute post ergocycle exercise

	T0	T1	T2	p	Post hoc
Coagulation parameter					
Activated partial thromboplastin time (second)	26.4 ± 0.5	26.8 ± 0.4	27.7 ± 0.4	0.01*	T2 > T0, T2 > T1
Prothrombin time (second)	10.2 ± 0.3	10.5 ± 0.3	10.7 ± 0.2	0.03*	T2 > T0
International normalized ratio	0.88 ± 0.02	0.90 ± 0.02	0.97 ± 0.03	0.01*	T2 > T0
Platelet [thousand/ml]	262 ± 25	252 ± 24	249 ± 25	0.02*	T2 < T0
Systemic inflammation					
Erythrocytes sedimentation rate 1 [ml/hr]	27.0 ± 11.3	25.4 ± 9.3	30.4 ± 11.1	0.07#	–
Erythrocytes sedimentation rate 2 [ml/hr]	47.3 ± 13.8	49.7 ± 12.4	50.4 ± 15.6	0.63	–
Blood glucose [mg/dL]	202 ± 35	173 ± 33	158 ± 30	< 0.01*	T2 < T1 < T0

#Greenhouse-Geiser correction; *significant; T0 — baseline; T1 — within 5 minutes after ergocycle exercise; T2 — 30 minutes after ergocycle exercise

study protocol was well-tolerated by middle-aged and older adults with T2DM. To our best knowledge, this present study is the first to report the effect of moderate-intensity ergocycle exercise on clinical coagulation parameters and to explore the association of the coagulation parameters with systemic inflammation and blood glucose among T2DM patients.

The most important clinically relevant findings of this present study were the elevation of aPTT and PT and the reduction of platelet counts that were observed after the short-term moderate-intensity ergocycle exercise, thus, indicated a tendency toward the improvement of coagulation. The findings were in consonance with a study that reported increases in aPTT, PT and fibrinolysis activities after vigorous exercise among diabetes patients [11]. However, the finding was in contrast with the increases in pro-coagulant activity that were reported among individuals with non-T2DM patients after short-term strenuous exercise testing [12]. Similarly, another study also reported the increased risk of thromboembolic incidents in running athletes after a prolonged vigorous exercise session [18]. These conflicting findings support the notion that the effect of exercise on the coagulation system varies and depends upon the exercise type, intensity, duration as well as different subjects' medical conditions [13].

Although there were some variations of findings across previous studies, the absence of the transient hypercoagulation after a short moderate-intensity exercise in this present study was a significant finding because it highlights the safety aspect of the exercise among T2DM patients. The finding, however, needs to be interpreted with caution. The coagulation parameters of this study participants were still within the normal range throughout the study period. Thus, the increases of aPTT and PT as well as the reduction of platelet count in this study, although were statistically significant, may not be clinically significant. Also, the coagulation responses to the exercise on T2DM patients with impaired coagulation system may differ from our study participants with normal coagulation parameters. Future studies, thus, need to further confirm the effect of the exercise on T2DM patients with impaired coagulation parameters.

The increase of erythrocyte sedimentation rate (ESR), one of the systemic inflammation markers, among individuals with T2DM population has been reported [19], thus, the high baseline ESR values in this present study were expected. This present study, however, failed to demonstrate an improvement of ESR after a short-term moderate-intensity exercise. The finding was in contrast with a recent study that reported improvements in ESR in individuals who engaged in regular moderate physical activity levels [20]. Chronic physiological adaptation may be required for ESR improvement. Also, the ESR is a non-specific inflammation marker that could be affected by conditions unrelated to T2DM such as aging, infections or malignancies [20]. Future studies, thus, should consider investigating the effect of both short- and long-term exercise programs on the systemic inflammation and take into account systemic inflammation markers mostly associated with T2DM conditions such as interleukin-6 and C-reactive protein [21].

Our study also demonstrated steady and significant improvement in blood glucose throughout the study period that was in line with the body of evidence that shows the positive effect of exercise in improving blood glucose levels [22, 23]. It has been recognized that exercise increases blood glucose utilization through the increased activity of the glucose transporter-4, which subsequently promotes the active muscle's glucose uptake to supply the fuel for muscular activity [24]. The exercise also increases cell insulin sensitivity, which

stimulates blood glucose uptake via a separate channel [24]. These cumulative events eventually reduce the level of circulating blood glucose after the exercise.

Lastly, although the blood glucose improvement in this present study was clinically significant, after 30 minutes of recovery time, the blood glucose was still above the recommended level of 110 mg/dL [25]. Since further observations were not made after the 30-minute period, a conclusion could not be drawn whether the blood glucose would continue to decrease and eventually achieved the recommended level after the study period. Herein, further studies are recommended to utilize the use of continuous glucose monitoring to confirm the magnitude, trend and duration of blood glucose improvement after exercise interventions are recommended. The information resulted from those studies will be required for assisting the T2DM patients in adjusting their exercise regimen and for guiding clinicians in prescribing anti-diabetic medication to achieve optimal glycemic control while avoiding hypoglycemia.

The key strength of this study was the utilization of the recommended exercise principles for T2DM patients [25]. Thus, findings from this study could be directly applied to T2DM populations. This study has confirmed the feasibility aspect of the moderate-intensity ergocycle exercise. The study also highlighted the safety aspect of the exercise and confirmed its potential role in improving hypercoagulation and blood glucose among middle-aged and adults with T2DM. The findings from this present study, however, must be interpreted in the context of certain limitations. First, this study was intended to assess the feasibility of the exercise protocol and only used a small sample size, thus, it may have resulted in inadequate statistical power for detecting changes and associations in all parameters measured in this study. However, effect sizes could be approximated from this study for calculating the appropriate sample size for future studies. Second, the values of the coagulation parameters were still within the normal range at baseline, thus the findings of this study may not be generally applied for T2DM patients with impaired coagulation. Third, this study only explored the coagulation parameters and did not include the fibrinolytic parameters such as tissue plasminogen activator, plasminogen activator inhibitor-1, and d-dimer. For a complete interpretation of coagulation and fibrinolytic activities, future research is recommended to include these parameters. Last, the ESR that was used in this study is a nonspecific systematic inflammation marker. Thus, the use of systemic inflammation markers more related to T2DM pathogenesis such as CRP and IL-6 is recommended for future research.

Conclusions

This study has shown that the short-term moderate-intensity ergocycle exercise was well tolerated by middle-aged and older T2DM patients and resulted in improvements in coagulation parameters and blood glucose levels. The systemic inflammation characterized by the erythrocyte sedimentation rates, however, was unaffected. Future studies are recommended to investigate the chronic adaptation of the coagulation parameters after ergocycle training among T2DM patients with coagulation impairments.

Conflicts of interest

The authors declare to have no conflict of interest.

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A new scoring system to predict the incidence of new onset diabetes after transplantation (NODAT)

ABSTRACT

Background. We performed this study to develop a new scoring system to stratify different levels of risk of developing new onset diabetes after transplantation (NODAT) in patients who underwent renal transplantation. Many prognostic variables have been previously described but few efforts have been made to group them in order to enhance their individual predictive power.

Material and methods. In a first phase, 100 patients were prospectively analysed to determine which factors were significantly associated with the development of NODAT. A risk score ranging from 0 to 10 points was developed using a multivariate analysis. In a second phase, such score was validated in a new sample of 100 patients.

Results. BMI ≥ 23.5 kg/m^2, age ≥ 38.5 years, fasting blood sugar at 1st post-operative day ≥ 159.5 mg/dL, fasting blood sugar at 5th post-operative day ≥ 122.5 mg/dL and HOMA-IR ≥ 2.5 were found as independent prognostic variables. A clear distinction was shown among categories of low, intermediate and high risk, defined according to the risk score.

Conclusion. This new scoring framework is basic and simple to accomplish. It permits a generally excellent stratification of risk of developing NODAT in patients undergoing renal transplantation. They might be separated in three risk stratification cohorts, which could be of help in early identification of NODAT. (Clin Diabetol 2020; 9; 4: 226–232)

Key words: NODAT, risk score, India, renal transplantation

Introduction

New-onset diabetes after transplantation (NODAT) refers to diabetes that occurs in previously non-diabetic persons after solid-organ transplantation, according to International consensus guidelines published in 2003 [1, 2]. There are many risk factors of NODAT. Some risk factors are the same as in general risk factors for diabetes mellitus (DM), while some others are specific to transplantation.

Some common risk factors include age, obesity, African-American and Hispanic [3–6]. In addition, some risk factors are unique to the transplant population. These include specific agents used for immunosuppression, human leukocyte antigen mismatch, donor sex and type of underlying renal disease [7]. Impaired glucose tolerance prior to transplant [8] and hyperglycaemia in the immediate perioperative period [9, 10] may identify patients at higher risk for the development of NODAT. There is paucity of data with regards to the development of risk scores for the development of new onset diabetes after transplantation (NODAT) from South-East Asian population. Although data from western population are available, we intended to develop the same for our population which is place of residence for almost one-fifth of the world's population. Furthermore, the pre-transplant and peri-transplant

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risk factors in our study population also differ considerably from the western population which calls for development of population specific predictive model for development of NODAT in the intended population. We conducted the present study to test the prognostic value of a combination of such risk factors resulting in a prospectively designed score that could be capable of making a clear distinction of different clinical outcomes with regards to development of NODAT applied to patients coming to hospital for renal transplantation. With that purpose we chose the most widely available prognostic variables that, in our model, provided the best independent information for the development of NODAT. The new score was applied in another cohort of patients consecutively admitted to renal transplant units who were not enrolled in trials of therapeutic interventions.

Material and methods

Study population

This was a single-centred prospective study of 200 subjects who underwent renal transplantation over a period of four years in a tertiary care centre in eastern India.

The inclusion criteria comprised of adult subjects with end stage renal disease who underwent live donor kidney transplantation and absence of diabetes prior to kidney transplantation, defined according to the American Diabetes Association (ADA) guideline. None of these patients were on any oral hypoglycaemic agents or insulin prior to kidney transplantation. All patients received their allograft from a living (related or unrelated) donor. All subjects received standard immunosuppressive medications that included triple immunosuppressive medications namely tacrolimus, mycophenolate mofetil or mycophenolate sodium and steroids with induction (ATG). Immunosuppressive therapy comprised tacrolimus (initiation dose of 0.15 mg/kg) (with target blood level of tacrolimus 10–15 ng/ml between 1st 3month, 5–10 ng/ml between 3rd to 6th month and 3–5 ng/ml in 6th to 12th month), prednisolone (20 mg/d) (with gradual tapering of dose 2.5 mg per month with target of 5 mg at the end of 6th month), and or mycophenolate mofetil (1.5 g/d). Previous studies in NODAT have modified the immunosuppressive regimen to prevent NODAT development. But ADA recommends that immunosuppressive regimens associated with best patient and graft survival should be used, irrespective of post-transplantation diabetes mellitus risk [11]. Subjects who were capable of understanding the study and gave informed written consent for study participation were only included. Patients with a diagnosis of DM prior to kidney transplantation based on ADA criteria, for diagnosis of DM [12], or those receiving anti-diabetic medications or those who were not capable of providing consent were excluded from the study.

'Prediabetes' in our study was defined according to ADA 2016 guidelines as HbA_{1c} value 5.7–6.4%. Those who are non-diabetic and underwent renal transplantation are further evaluated for the development of NODAT during 1-year post-transplantation follow-up. Post-transplant follow-up done on weekly basis for 1st month, every 15th day from 1st month to 3rd month, monthly from 3rd month to 12th month. Each transplant patient was followed up for 1-year post-transplant or for 6 months post-development of NODAT, whichever is later. NODAT was defined according to standard ADA criteria provided the patient was receiving therapy (oral hypoglycaemic drugs or insulin) at 3 months post-transplant. Immediate posttransplant hyperglycaemia was defined as a random blood sugar (RBS) $\geq$ 200 mg/dL [1] or requirement of insulin on $>$ 2 days whereas the patient was of dextrose-containing fluid infusions (usually from the 4th postoperative day).

In addition to routine transplant workup, pretransplant BMI, family history of DM, HbA_{1c}, fasting insulin level, fasting C-peptide level, serology for hepatitis B, C and serum magnesium level were evaluated in all patients 2 days prior to transplant. Pre-operative insulin resistance (HOMA-IR), insulin sensitivity (HOMA-S) beta-cell function (HOMA-B and C-peptide levels) were assessed. All the above pre-transplant variables were further compared between NODAT and non-NODAT subjects at the end of the study to assess their strength of association.

Data management and statistical analysis

Descriptive statistics was analysed with SPSS version 17.0 software for windows. In order to develop a risk score, all demographic, clinical, and biochemical variables were routinely collected. Continuous variables were presented as mean ± SD and analysed by unpaired t test. Categorical variables were expressed as frequencies and percentages. Nominal categorical data between the groups were compared using Chi-square test or Fischer's exact test as appropriate. A p value of $<$ 0.05 was considered to be statistically significant. Univariate was analysis done to evaluate odds ratio of various parameters associated with increased risk of NODAT among study population.

Every variable resulting in a p value $<$ 0.01 in the univariate model was entered into a multiple logistic regression analysis to determine which were independently related to the end-points.

The predictive accuracy of the multivariate model was evaluated using the C statistic, an index that

Table 1. Baseline characteristics of the subjects who developed NODAT in development phase and validation phase

	Development phase (n = 100)	Validation phase (n = 100)
Age (years), mean (SD)	45.2 (10.93)	46 (11)
Family H/O diabetes mellitus, n (%)	25 (25)	32 (32)
BMI [kg/m^2], mean (SD)	22.62 (4.03)	23.15 (5.77)
Hepatitis B infection, n (%)	3 (3)	3 (3)
Hepatitis C infection, n (%)	3 (3)	3 (3)
Autosomal dominant polycystic kidney disease, n (%)	4 (4)	3 (3)
Mean magnesium levels [mEq/L], mean (SD)	1.84 (0.51)	1.88 (0.37)
Mean total cholesterol levels [mg/dL], mean (SD)	139.41 (35.01)	136 (21)
Mean triglyceride levels [mg/dL], mean (SD)	84.02 (64.52)	89.5 (57.21)
Pre-operative HbA_{1c} > 5.7%, n (%)	14 (14)	16 (16)
Pre-operative HbA_{1c} (%), mean (SD)	5.34 (0.16)	5.12 (0.1)
ABO compatibility transplant, n (%)	94 (94)	96 (96)
HOMA-IR, mean (SD)	1.87 (1.08)	1.84 (1.11)
HOMA-S, mean (SD)	79.35 (48.07)	84.57 (40.18)
HOMA-beta cell function, mean (SD)	64.14 (3.64)	68.23 (4.21)
C-peptide level, mean (SD)	11.06 (5.09)	10.21 (6.21)

Table 2. The results of the univariate analysis

Variable	OR	95% CI	P value
Age (years), mean ± SD	1.084	1.033–1.138	0.004
Family H/O diabetes, n (%)	1.133	1.013–1.890	< 0.001
Fasting blood sugar-day 1 [mg/dL]	2.032	1.412–3.785	< 0.001
BMI [kg/m^2], mean ± SD	1.363	1.178–1.577	< 0.001
Hepatitis B infection, n (%)	7.4	0.638–85.81	0.28
Hepatitis C infection, n (%)	7.4	0.638–85.81	0.28
Fasting blood sugar-day 5, mean ± SD	9.28	5.413–16.519	< 0.001
Autosomal dominant polycystic kidney disease	1.001	0.856–1.087	0.242
Mean magnesium level [mEq/L], mean ± SD	0.780	0.315–1.930	0.305
Mean total cholesterol level [mg/dL], mean ± SD	1.015	0.997–1.032	0.120
Mean triglycerides level [mg/dL], mean ± SD	1.008	0.998–1.018	0.066
Pre-operative HbA_{1c} (%) > 5.7%	2.315	1.389–2.561	< 0.001
Pre-operative HbA_{1c} (%), mean ± SD	1.057	1.029–1.185	0.001
ABO compatibility transplant	0.135	0.023–0.792	< 0.001
HOMA-IR	0.987	0.932–0.998	0.001
HOMA-S	0.957	0.921–0.997	< 0.001
HOMA-beta cell function	0.956	0.901–0.978	0.001
C-peptide level	1.987	1.057–2.184	< 0.001

reflects the area under the receiver operating characteristic curve.

The odds ratio (OR) values obtained in the multivariate analysis were used to develop the scoring system in the following way: if the OR was between 1 and 1.9, one point was adjudicated; two points if it was between 2 and 2.9; three points between 3 and 3.9 and four points if it exceeded the last value.

Once the risk score was developed, we conducted a validation phase to assess its prognostic accuracy in a prospectively collected new sample of patients.

The overall predictive ability of the risk score was then assessed with the C statistic and compared with that obtained from the multivariate model of the development phase.

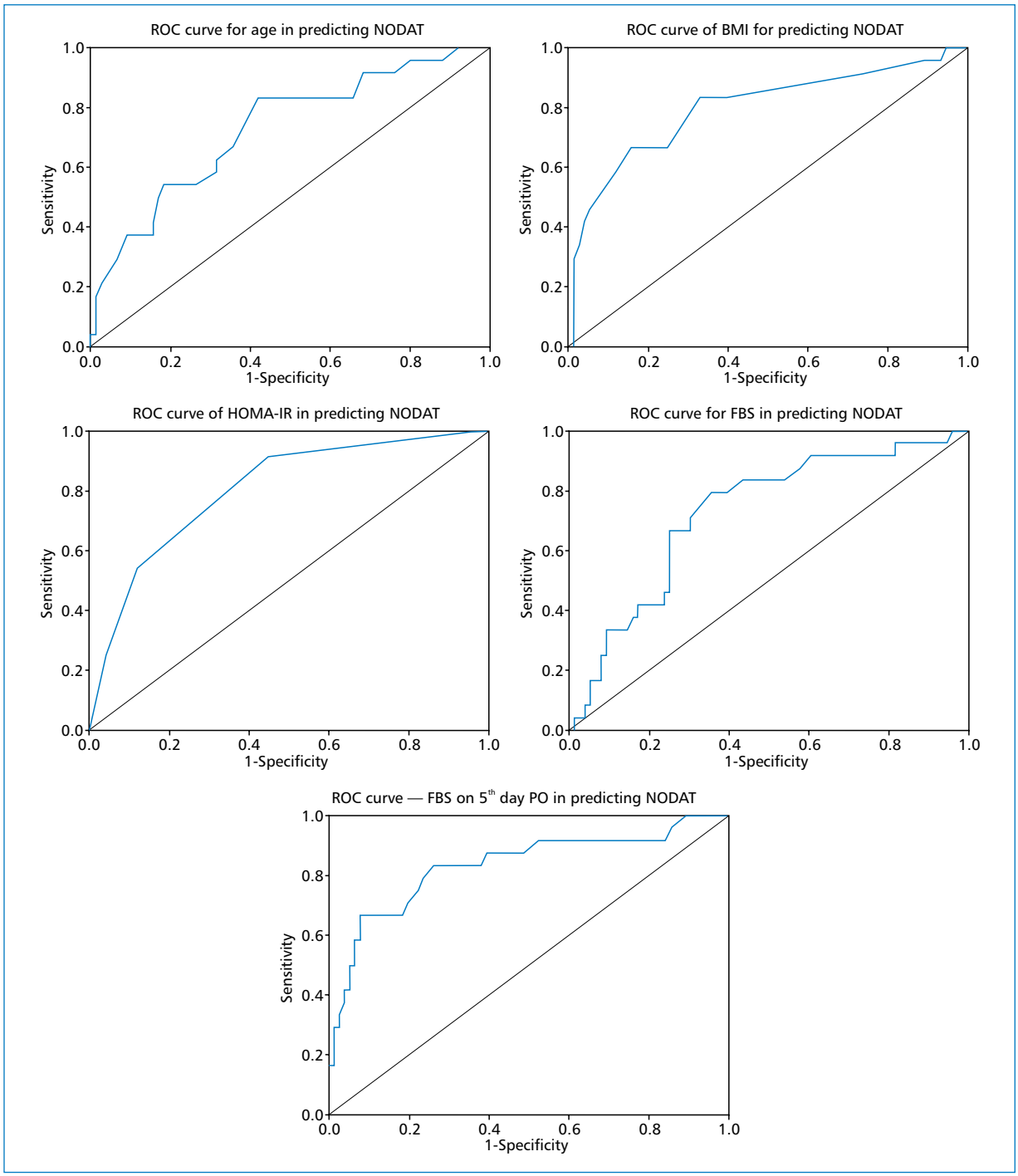

Figure 1. ROC of the significant predictor variables

Results

Score development phase

One hundred patients were prospectively included in this phase. Among the 100 subjects included in the analysis, 24 patients (19 males and 5 females) developed NODAT during 1 year of follow-up after transplantation.

Eighteen variables were included in the univariate analysis. We included the variables which had a p value less than 0.010 for multivariate analysis. Hence, we included 14 variables in the multiple regression model and only BMI, age, fasting blood sugar at 1st and 5th post-operative day and HOMA-IR were found as independent prognostic variables of NODAT. The C

Table 3. Results of the multivariate analysis

	B — coefficient	C — statistics	Best cut-off	Sensitivity	Specificity	OR, 95% CI
Age	0.703	0.76	38.5	83.33	71.4	1.231 (1.133–1.938)
BMI	0.821	0.80	23.5	75.6	84.2	2.103 (1.278–2.878)
HOMA-IR	1.439	0.81	2.5	74.2	78.8	4.062 (2.79–5.81)
FBS-1st day POD	0.694	0.72	159.5	77.6	82.9	1.011 (1.002–1.018)
FBS-5th POD	0.936	0.84	122.50	79	92.1	2.082 (1.037–4.088)

Table 4. Outcomes according to risk categorization in the validation phase

	Low risk (n = 46)	Intermediate risk (n = 26)	High risk (n = 28)
Incidence of NODAT	4 (8.7)	9 (34.62)	13 (46.43)
OR for high *vs.* low risk		5.34, 95% CI 2.9–11.8	
OR for high *vs.* intermediate risk		1.34, 95% CI 1.169–9.82	
OR for intermediate *vs.* low risk		3.98, 95% CI 2.1–6.7	

statistic for the multivariable model was 0.79 (95% CI 0.74–0.89).

Development of scoring system

Therefore, according to the OR obtained, the scoring system was established as follows:

— age ≥ 38.5 years (OR = 1.231): 1 point;
— BMI ≥ 23.5 kg/m^2 (OR = 2.103): 2 points;
— HOMA-IR ≥ 2.5 (OR = 4.062): 4 points;
— FBS on 1st POD ≥ 159.5 (OR = 1.011): 1 point, and
— FBS 5th day POD ≥ 122.5 (OR = 2.082): 2 points.

As the highest possible score was 10 points, we divided it in tertiles so that we could assign each patient to one of three categories according to the score sum value: low-risk when it was 0 to 2, intermediate-risk when it was 3 to 6 and high-risk when it was 7 to 10.

Validation phase

One hundred patients entered this phase of the study. Baseline characteristics were similar to the first except for a slightly higher prevalence of family history of diabetes in the validation cohort. The incidence of NODAT was similar in the development phase (24%) and validation phase (26%).

The incidence of NODAT occurred in 8.7% of low risk patients, 34.62% of intermediate risk and 46.43% of high risk patients (OR for high *vs.* low risk: 5.34, 95% CI 2.9–11.8, P < 0.001; OR for high *vs.* intermediate risk: 1.34, 95% CI 0.69–9.82, P = 0.044; OR for intermediate *vs.* low risk: 3.98, 95% CI 2.1–6.7, P = 0.001).

Predictive power of the score, as assessed by the C statistic was 0.78 (95% CI 0.67–0.88), similar to that found for the multivariate model.

Discussion

Although many demographic, clinical and biochemical markers have been clearly shown to correlate with development of NODAT, few efforts have been made to group them in order to improve their individual predictive power.

The scoring system proposed here is quite simple to implement and has a good ability to discriminate risk according to the C-statistic value. All the information needed in our study to predict NODAT namely age, BMI, HOMA-IR and FBS are easily available. All of these are non-expensive and most importantly, they have a very good prognostic value. We divided the population studied into three groups: low, intermediate and high risk, which is a common practice among clinicians regarding many chronic diseases.

In the present study, age > 38.5 years was found to be a significant predictor for the development of NODAT. Increasing age is associated with increased risk for NODAT aspecially over the age of 40 years [6, 10, 12]. A study by Crosio et al. in 2078 patients showed that patients older than 45 years were 2.9 times increased risk of developing diabetes [13]. Every 10-year increase in age leads to 1.5-fold increased risk of diabetes [14].

In our study, pre-transplant BMI > 23.5 kg/m^2 is found to be significant prognostic variable for development of NODAT. Obesity independently correlates with the development of NODAT [3–5, 15, 16]. An analysis of 15,309 patients using the Organ Procurement and Transplant Network/United Network for Organ Sharing (OPTN/UNOS) database found that the risk of NODAT increased 1.4-fold for those with a BMI of 25–30 and nearly doubled if the BMI was > 30 [17]. It remains unclear whether weight gain after transplantation is

the cause, however one study suggested pre-transplant weight increases the risk for NODAT [18].

Midtvedt et al. using hyperinsulinaemia euglycemic clamps found insulin resistance as a common denominator of KTRs with NODAT and IGT [19]. In study done by Bayes et al. NODAT patients showed significantly higher pre-transplant plasma insulin concentrations and HOMA-IR index compared to non-NODAT patients [20]. We found HOMA-IR of more than 2.5 had an OR of 4.062 for NODAT development.

Patients with post-transplant hyperglycemia in our study had a fourfold higher risk of developing NODAT. Similar results were seen in the study by Chakkera et al. in 200 posttransplant patients in Arizona [21]. A study from Chile reported 5.4-fold higher risk of developing diabetes in patients with early hyperglycemia [22]. A French study found first post-transplantation capillary blood glucose and fasting blood glucose on 1st day tended to be higher in patients who developed diabetes 3 months later [23]. They reported maximum hyperglycaemia on the first post-operative day which decreased gradually during first 4 days of transplantation probably related to decrease in corticosteroid dosages and reduction in insulin resistance due to resolution of uraemia. A Belgian study demonstrated that a normal OGTT on the 5th post-operative day was associated with a significantly decreased risk of NODAT at 3 months [24]. We also found that persistent post-operative day 5th FBS to have a higher odd of NODAT development compared to day 1 FBS. The significant risk of NODAT posed by posttransplant hyperglycaemia makes it prudent to follow up these patients more diligently and are likely to benefit from intensive glucose monitoring. Based on the available evidence, NODAT cannot be efficiently prevented by tailored immunosuppression alone without compromising kidney graft survival. In the TIP-study, early use of basal insulin in the immediate post-transplantation period lower odds of NODAT by 73% throughout 1 year of follow-up [25].

Only one previous study in predominantly white transplant recipients described a pretransplant predictive risk model for NODAT using seven pretransplant variables (age ≥ 50 years, planned use of maintenance corticosteroids; use of gout medicine; BMI ≥ 30 kg/m^2; fasting glucose ≥ 100 mg/dL; fasting triglycerides ≥ 200 mg/dL; and family history of type 2 diabetes) [26]. But they did not consider peri-transplant risk factors like immediate post-operative hyperglycemia which are strongly associated with NODAT. Moreover, there is variability in the use of gout medication and corticosteroids. In our study, be had a standard immunosuppressive regimen for best transplant outcome in all patients and according to OR obtained, scoring system using five variables (age ≥ 38.5 years; BMI ≥ 23.5 kg/m^2; HOMA-IR ≥ 2.5; FBS on 1st POD ≥ 159.5; FBS 5th day POD ≥ 122.5) was established. There is always a concern of higher mortality in the person who develops NODAT which warrants adaptation of risk scores in routine clinical setting by transplant physicians. A study by Cooper et al. with 266 participants undergoing kidney transplantation found the age and sex adjusted mortality to be 1.69 times higher among patients with NODAT versus those without NODAT, hazard ratio 2.69 (95% CI 1.04–7.01) [27]. Cosio et al. described two fold increase in mortality with NODAT compared to nontransplant recipients, which was equal to that of pretransplant diabetes and independent of other factors known to reduce survival [28].

Conclusion

The morbidity and mortality associated with NODAT makes it prudent to a identify risk factors and develop a risk score for early detection of NODAT and stratify effective strategies for prevention and intensive treatment in resource-limited setting wherein extensive monitoring in all patients is expensive. This new scoring framework is basic and simple to accomplish. It permits a generally excellent stratification of risk of developing NODAT in patients undergoing renal transplantation. They might be separated in three risk stratification cohorts, which could be of help in the decision-making process. Our findings should be tested in a larger cohort of patients in order to suggest clinical strategies based on them. If these data were confirmed, a highly aggressive approach as per recent evidences can be recommended in high risk and intermediate risk patients and a more conservative one could be reserved for the low risk group.

Conflict of interest

The authors declare to have no conflict of interest.

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Clinical effectiveness of combination therapy with dulaglutide, SGLT2 inhibitor and metformin with or without insulin in Indian adults with type 2 diabetes: a real-world retrospective study

ABSTRACT

Background. Antidiabetic agents such as glucagon-like peptide-1 receptor agonists (GLP-1RAs) and sodium glucose cotransporter-2 inhibitors (SGLT2i) are known to improve glycaemic control with favourable impact on cardiovascular (CV) risk factors. Given the potential benefits, this study evaluated the clinical effectiveness of combination therapy including dulaglutide, SGLT2i and metformin with or without insulin in Indian adults with inadequately controlled T2DM.

Material and methods. This retrospective, real-world, single-centre study included 15 adults (mean age [SD, standard deviation]: 49.47 [9.29] years) with inadequately controlled T2DM. The patients received a combination of dulaglutide, SGLT2i and metformin with or without insulin. Changes in fasting blood glucose (FBG), postprandial blood glucose (PPBG), glycosylated haemoglobin (HbA_{1c}), body weight, BMI, vascular age (VA), systolic blood pressure (SBP) and diastolic blood pressure (DBP) were examined for a follow-up duration of 3 months. Self-reported adverse events were also recorded.

Results. At the 3-month follow-up, the combination therapy resulted in significant reduction ($P < 0.001$) in glycaemic parameters such as FBG, PPBG and HbA_{1c} with a mean reduction (MR [SD]) of 66.67 (40.05) mg/dL, 83.33 (64.11) mg/dL and 1.78 (1.08) % respectively. A significant reduction ($P < 0.001$) in body weight and corresponding BMI was demonstrated with an MR (SD) of 6.40 (3.96) kg and 2.67 (1.63) kg/m^2 respectively. Similarly, significant reductions ($P < 0.05$) were also recorded for SBP (4.40 [4.61] mm Hg) and DBP (2.80 [3.84] mm Hg). The therapy was also associated with significant reduction ($P < 0.001$) in VA (MR [SD]: 3.93 [2.46] years). The therapy was well tolerated; however, self-reported gastrointestinal symptoms were reported in 5 patients, which subsided within 2 weeks of therapy initiation.

Conclusion. Combination therapy with dulaglutide, SGLT2i and metformin with or without insulin resulted in significant improvements in glycaemic parameters, body weight and SBP in Indian adults with inadequately controlled T2DM. Interestingly, there was a significant improvement in vascular age associated with the therapy. (Clin Diabetol 2020; 9; 4: 233–238)

Key words: dulaglutide, GLP-1RAs, India, SGLT-2i, T2DM

Introduction

Type 2 diabetes mellitus (T2DM) is considered a global epidemic affecting nearly 90% of population world-wide. Patients with T2DM are usually presented with comorbidities including hypertension, hyperlipidaemia, obesity and are therefore prone to cardiovascular (CV) risk [1]. Management of T2DM is complex given the progressive nature of the disease and associated

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co-morbidities. As the disease progresses, clinicians involved in diabetes care often tackle with the challenge of adjusting multiple medications in the face of renal failure and CV disease. In addition, medications have to be tailored to help patients overcome intolerances and adverse effects [2].

Newer generation glucose-lowering drugs, glucagon-like peptide-1 receptor agonists (GLP-1RA) and sodium glucose cotransporter-2 inhibitors (SGLT2i) have made a remarkable breakthrough into the T2DM treatment armamentarium enabling a patient centred approach. The complimentary mechanisms of action demonstrated by GLP-1RA and SGLT2i with respect to cardio and nephroprotective effects are generating their potential need and are currently attracting significant attention from medical research communities [3]. Cardiovascular outcome trials (CVOT) have demonstrated significant reductions in major adverse cardiovascular events (MACE) and mortality outcomes with GLP-1RA (LEADER, EXCEL, SUSTAIN-6) and SGLT2i (EMPA-REG, CANVAS) [4]. To further support this evidence, REWIND, the first CVOT trial for dulaglutide that included a majority of participants who did not have an established cardiovascular disease, is reported to have demonstrated superiority in the reduction of MACE [5]. Based on evidence from pivotal trials (DURATION-8, AWARD-10) and other recently published real-world observational studies, patients with poorly controlled T2DM treated with GLP-1RA as an add-on to SGLT2i have shown beneficial effects on glycaemic control, body weight and SBP with low hypoglycaemia risk compared with those assigned to placebo [1, 4, 6, 7]. A literature review on the efficacy of combined treatment with GLP-1RA and SGLT2i showed additive effect on lowering the HbA_{1c} levels, systolic blood pressure (SBP), diastolic blood pressure (DBP) and body weight in patients with T2DM. The combination therapy may therefore address multiple facets of the disease and may prove to be a potential additive in lowering cardiorenal risk in addition to glycaemic control [1, 8].

The aim of this retrospective, real-world, single-centre study is to evaluate the impact of combination therapy with once-weekly dulaglutide, SGLT2i and metformin with or without insulin on fasting blood glucose (FBG), post prandial blood glucose (PPBG), HbA_{1c}, body weight, body mass index (BMI), vascular age (VA), SBP and DBP in Indian patients with inadequately controlled T2DM in a real-world setting.

Materials and methods

Study design and participants

This retrospective, real-world, single-centre observational study was carried out at the AMRI Hospital, a tertiary care centre in Salt Lake, Kolkata, India between January, 2018 to March, 2018. Eligible patients (N = 15) were adults (mean age, 49.47 ± 9.29 years) with inadequately controlled T2DM. The mean duration of diabetes was 7.6 ± 1.24 years. All participants were receiving antidiabetic medications which included a full dose of gliptin (Sitagliptin [100 mg] or Vildagliptin [100 mg] or Linagliptin [5 mg]) and a full dose of Glimepiride (4 mg). Patients were stopped on these medications and shifted to a combination therapy with once-weekly dulaglutide (1.5 mg or 0.75 mg up-titrated to 1.5 mg), SGLT2i (canagliflozin [100 mg or 300 mg], empagliflozin [25 mg] or dapagliflozin [10 mg]) and metformin (500 mg or 1000 mg). Insulin glargine or degludec were subsequently required in 4 out of 15 patients to achieve a target FBS of 90–110 mg. The average dose of Basal Insulin was 8.35 ± 0.45 IU. Patients with type 1 diabetes, history of pancreatitis, family history of medullary thyroid cancer, or multiple endocrine neoplasia type 2 were excluded from this study. All patients were on statins and antihypertensives (to achieve individualised optimal target SBP and DBP) in addition to antidiabetic medications. Mean SBP 122.87 ± 4.81 mm Hg and mean DBP 84.27 ± 4.95 mm Hg.

Data analysis was conducted for patients, who were able to complete three months of therapy.

Endpoint assessment

Changes in clinical parameters including, FBG, PPBG, HbA_{1c}, body weight, BMI, VA, SBP and DBP were assessed at baseline and 3 months after treatment. AGEDIO B900 Pulse Wave Analysis System (Hibernia Medical). This instrument has a BP cuff that can be attached to the arms, a recording machine and a software which generates reports on the vascular age, based on the age, body weight, SBP and DBP and the arterial stiffness.

The report is generated in reference to the chronological age of the individual, i.e. same as or number of years more than or less than the chronological age.

Agedio is a screening service for arterial stiffness called VA measurement for the pharmacy store or other health care centers. The technology offers an (upper-arm) cuff-based blood pressure and arterial stiffness measurement within one single procedure. Arterial stiffness is quantified with the aortic pulse wave velocity (PWV), measured in m/s. Agedio consists of a validated measurement device and the displaying device, an iPad. It allows for a measurement within a few minutes and provides two detailed reports one for the patient and one for the physician. The medical report in addition to arterial stiffness includes the hemodynamic figures such as cardiac output, total vascular resistance and central (aortic) blood pressure.

Clinical indications for technology are as follows: Cardiovascular diseases such as coronary heart disease globally are the leading cause for morbidity and mortality. Risk assessment via blood pressure, LDL measurement or by using traditional risk scores such as Framingham and SCORE have shown to be insufficient for detecting the individual risk of a patient. The proportion of patients receiving inadequate treatment for cardiovascular diseases, such as high blood pressure, is alarmingly high. Existing measurement methods for blood pressure measurement do not provide enough medical information about the pathophysiological individual background of the patients.

Therefore, Agedio improves the current situation by following indications:

- leverage of patient awareness by using informative and catchy patient reports (pharmacy), supports the encouragement of patient therapy adherence;
- precise identification and classification of patients into low, medium and high risk patients (medical doctor);
- provision of hemodynamic figures for support in therapeutic decisions and thereby related clinical benefits, such as reduction of cardio-vascular events as strokes and myocardial infarctions (discovery).

The hemodynamic figures provide for a solid foundation of therapeutic decisions and benefits. They may provide the rationale for improved application for existing anti-hypertensive and other drugs. In comparison to regular therapy decision making, this advanced measure reduces emergency visits and re-hospitalization related to new cardiovascular and coronary events by 40%.

The added value of risk prediction of PWV, above and beyond traditional risk factors, including SCORE and Framingham, has been quantified in a number of studies. The net classification value (NRI, net reclassification index) of PWV measurement allows for both, patient re-classification into higher or lower risk categories. The results have a direct influence on therapeutic decisions.

Agedio is a main point of action for stakeholders involved in public health, focusing on services within the pharmacy. Do you know your vascular age is a concept intended for a long-term partnership between pharmacists, medical professionals, health insurance companies and patients.

Adverse events following therapy administration were also recorded.

Statistical methods

All statistical analysis was performed using R 3.4.1 and MS-Excel software. Comparison between baseline and follow-up value of various parameters was done using paired *t* test. Continuous variables were presented as mean (standard deviation [SD]) with 95% CI as applicable and categorical data were presented as proportions. All P values were 2-sided and were considered significant if $P < 0.05$.

Table 1. Baseline demographics of the study group

Variable	Baseline Mean (SD)
Age (years)	49.47 (9.29)
FBG [mg/dL]	201.67 (62.13)
PPBG [mg/dL]	248.20 (5.67)
HbA_{1c} (%)	8.61 ± 1.41
Body weight [kg]	98.60 ± 14.05
BMI [kg/m^2]	32.27 ± 4.67
VA (years)	53.73 ± 7.62
SBP [mm Hg]	129.87 ± 4.81
DBP [mm Hg]	84.27 ± 4.95

BMI — body mass index; CI — confidence interval; DBP — diastolic blood pressure; FBG — fasting blood glucose; HbA_{1c} — glycosylated haemoglobin; LCL — lower confidence limit; PPBG — postprandial blood glucose; SBP — systolic blood pressure; SD — standard deviation; UCL — upper confidence limit; VA — vascular age

Results

Baseline demographics

A total of 15 (male: 9 and female: 6) patients received combination therapy with once-weekly dulaglutide, SGLT2i and metformin with or without insulin for 3 months. Baseline characteristics of the study population are presented in Table 1.

Impact on clinical outcomes

Improvement in clinical outcomes with dulaglutide as add-on therapy is presented in Table 2.

Glycaemic parameters

A significant decline ($P < 0.001$) in FBG, PPBG and HBA_{1c} from baseline with a mean reduction (MR) of 66.67 mg/dL, 83.33 mg/dL and 1.78% respectively was demonstrated at 3-months follow-up.

Non-glycaemic parameters

Body weight

The therapy demonstrated a significant reduction ($P < 0.001$) in body weight (MR: 6.40 kg) and corresponding BMI (MR: 2.67 kg/m^2) at 3-month follow-up.

Vascular age

The therapy was associated with a significant reduction ($P < 0.001$) in VA (MR: 3.93 years) at the follow-up.

Table 2. Improvement in outcomes from baseline to follow-up in the overall population

Variable	Mean improvement	Standard deviation	95%CI (LCL, UCL)	P value
Glycaemic parameters				
FBG [mg/dL]	66.67	40.05	(46.40–86.93)	< 0.001
PPBG [mg/dL]	83.33	64.11	(50.89–115.78)	< 0.001
HbA_{1c} (%)	1.78	1.08	(1.24–2.33)	< 0.001
Non-glycaemic parameters				
Body weight [kg]	6.40	3.96	(4.40–8.40)	< 0.001
BMI [kg/m^2]	2.67	1.63	(1.84–3.49)	< 0.001
VA (years)	3.93	2.46	(2.69–5.18)	< 0.001
SBP [mm Hg]	4.40	4.61	(2.07–6.73)	< 0.05
DBP [mm Hg]	2.80	3.84	(0.86–4.74)	< 0.05

BMI — body mass index; CI — confidence interval; DBP — diastolic blood pressure; FBG — fasting blood glucose; HbA_{1c} — glycosylated haemoglobin; LCL — lower confidence limit; PPBG — postprandial blood glucose; SBP — systolic blood pressure; SD — standard deviation; UCL — upper confidence limit; VA — vascular age

Blood pressure

Follow-up at 3-months demonstrated a significant reduction ($P < 0.05$) in SBP (MR: 4.40 mm Hg and DBP (MR: 2.80 mm Hg).

Adverse events

Self-reported gastrointestinal symptoms were observed in 5 (33.33%) patients which included nausea and bloated abdomen. All adverse events subsided within 2 weeks of therapy initiation. Pantoprazole and domperidone combination were administered in 3 out of the 5 patients to mitigate the gastrointestinal side effects. No cases of severe hypoglycaemia were reported.

Discussion

Patients with T2DM often present metabolic and cardio-renal co-morbidities, adding complexity to diabetes care. With the advent of newer drugs such as GLP--1RA and SGLT2i, the diabetes treatment is undergoing a paradigm shift towards improved diabetes care [7]. The current study evaluated the clinical effectiveness of combination therapy with once-weekly dulaglutide, SGLT2i and metformin with or without insulin in Indian adults with T2DM.

This study demonstrated significant reductions in glycaemic parameters such as FBG, PPBG, HbA_{1c} for a follow-up period of 3 months. In addition, a significant reduction in body weight and BMI was observed. This is supported by one of the pivotal clinical trials for dulaglutide, AWARD-10 which compared the efficacy of combination therapy of dulaglutide and SGLT2i versus placebo and SGLT2i in patients with T2DM. Patients who received dulaglutide as an add-on to SGLT2i showed statistically superior improvements in glycaemic control compared with placebo and SGLT2i. Similarly changes in weight, FBG and SBP was greater for the dulaglutide arms compared with placebo [9]. This finding is also in-line with another real-world study wherein the effectiveness of co-administration of GLP-1RA and SGLT2i was evaluated [7]. Notably, in a retrospective observational study by Curtis et al., which assessed the effect of combination of add-on therapy of GLP-1RA and SGLT2i on glycaemic control and weight in T2DM patients, reported significantly larger reduction in HbA_{1c} with dual therapy (GLP-1RA and an SGLT2i) compared with a diabetic regimen with GLP-1RA alone. This effect was sustained along with the additional benefit of further weight loss in 58% of the patients on dual add-on therapy compared to single GLP-1 therapy [10].

Clinically relevant improvement in daily and long-term glucose control with combined use of dulaglutide and SGLT2 inhibitors is important in the context of the differences in mechanisms of actions between GLP-1 receptor agonists and SGLT2 inhibitors — enhancement of insulin secretion and glucagon suppression with GLP-1 receptor agonists versus increased urinary glucose excretion and glucagon stimulation with SGLT2 inhibitors. Understanding the effect on glucagon was of special interest in this study because treatment with SGLT2 inhibitors has been shown to increase glucagon concentrations, potentially interfering with glucagon-lowering actions of GLP-1 receptor agonists. Importantly, both dulaglutide doses suppressed serum glucagon secretion, with dulaglutide 1.5 mg achieving a significantly greater reduction from baseline versus placebo, consistent with the reductions in fasting glucagon concentration of similar magnitude reported in other studies of dulaglutide. The preserved effect of dulaglutide on glucagon when added to SGLT2 inhibi-

tor treatment might be an important contributor to the overall glucose-lowering potency in this patient This preserved effect is not seen with dipeptidyl peptidase-4 (DPP-4) inhibitors or liraglutide when added to treatment with SGLT2 inhibitors since mean glucagon concentrations remained unchanged. This finding is potentially due to the lower potency of DPP-4 inhibitors and liraglutide compared with dulaglutide in the suppression of glucagon secretion [9]. The reduction in HbA_{1c} and bodyweight with dulaglutide 1.5 mg in our study. One possible reason could be related to the simulteneous introduction of SGLT2 inhibitor therapy in patients on a background of medications which were weight neutral. Continuous decrease in HbA_{1c} and bodyweight suggest that the bodyweight-lowering action of simulteneously introduced SGLT2 inhibitors was ongoing during the treatment period.

A significant reduction in SBP and was noted at the 3-month follow-up. This is in contrast with a real-world study by Ghosal et al. which assessed the effectiveness of combination therapy with dulaglutide and SGLT2i in Indian patients with T2DM. However, the results are in-line with clinical trial, AWARD-10 as elaborated above and other retrospective studies [7, 9].

Our study for the first time, reported the changes in vascular age as a result of combination therapy with dulaglutide and SGLT2i. Vascular age reflects the real atherosclerotic damage and is considered as a new variable in cardiovascular risk assessment [11, 12]. This study demonstrated significant improvement in vascular age at the follow-up compared with baseline. This is important as the assessment of cardiovascular risk is a cornerstone in effective prevention and management of cardiovascular disease often associated with T2DM.

Combination therapy was well tolerated; however, self-reported gastrointestinal symptoms subsided within 2 weeks of therapy initiation. There were no cases of severe hypoglycaemia reported in this study.

Study limitations

The most important limitation of this study is lack of a comparator arm. This leaves the possibility of risk of reporting bias that may affect the results. In addition, the study duration was relatively short (3 months). This study has a retrospective, single-centre observational design and small sample size. In an Indian setting, one of the barriers in accessing medications including GLP-1RA includes cost which limits the exposure of the drug to a narrow pool of patient population. Thus, these results might not be reflective of diverse patient population adding to selection bias. However, the importance of this study was not significantly affected by these limitations.

Conclusion

In a real-world setting, co-administration of dulaglutide with SGLT2i and other antidiabetic medications demonstrated significant improvements in glycaemic parameters and SBP. Interestingly, for the first time, the study reports improved vascular age with the therapy. The therapy was well tolerated with no serious adverse events reported. This study therefore provides a pilot and indeed generates a hypothesis supporting combination of GLP-1RA, SGLT-2i and other antidiabetic drugs, in the management of T2DM, which needs to be tested in a more systematic and appropriately powered study design. Further research is warranted in terms of healthcare resource practice, costs and long-term outcomes.

Compliance with ethics

All procedures were followed in accordance with the responsible committee on human experimentation and with the Helsinki Declaration of 1975 and subsequent revisions, and informed consent was received from the patient involved in this study.

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Conflict of interest

The authors declare to have no conflict of interest.

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Serum leptin level and microvascular complications in type 2 diabetes

ABSTRACT

Background. Type 2 diabetes (T2DM) and its complications are highly prevalent in Egypt and are considered a major health problem. Insulin resistance arising from visceral obesity is the main pathological mechanism of T2DM. Leptin is an adipokine secreted from visceral adipose tissue and its level is proved to be higher in patients with T2DM, but its association with microvascular complications is not yet well-established, for this aim the present study was conducted.

Methods. This cross-sectional study was conducted among 120 participants with T2DM recruited from the diabetes outpatient clinic of Alexandria Main University Hospital, Alexandria, Egypt. Each participant was subjected to full history taking, complete physical examination and laboratory investigations.

Results. Serum leptin level was significantly positively correlated with diabetes duration, BMI, WC, systolic and diastolic blood pressure, FPG, HbA_{1c}, serum insulin level, HOMA-IR, total cholesterol, triglycerides and LDL-C. Regarding microvascular complications, serum leptin level was highly significantly positively correlated with UACR, peripheral neuropathy and retinopathy ($P < 0.001$) and significantly negatively correlated with e-GFR ($P = 0.003$).

Conclusions. Serum leptin level is significantly correlated with microvascular complications in patients with T2DM in Alexandria, Egypt. (Clin Diabetol 2020; 9; 4: 239–244)

Key words: leptin, type 2 diabetes, microvascular complications

Introduction

Type 2 diabetes (T2DM) represents 90–95% of patients with diabetes worldwide [1]. Egypt occupies the 8th ranks regarding the prevalence of diabetes. The estimated prevalence of diabetes in Alexandria, Egypt was 16.8% in 2018 [2]. The main pathogenic mechanism of T2DM is insulin resistance that results in relative insulin deficiency [1]. Most of patients with T2DM are obese and many studies linked obesity to T2DM [3, 4]. Insulin resistance in muscle and liver results from impaired glucose uptake by adipose tissue [5]. Visceral adipose tissue is metabolically active and secretes substances called adipokines. Adipokines leads to development of insulin resistance by many mechanisms [6, 7].

Leptin is one of the adipokines secreted from adipocytes in visceral adipose tissue [8]. It is also detected in gastric mucosa, hepatic stellate cells, placenta, ovaries and mammary gland [9]. Leptin is acting as an adipose-derived endocrine signal by inducing satiety through hypothalamic receptors and enhancing lipid metabolism and energy expenditure [10]. Higher serum leptin levels were detected in patients with diabetes and leptin resistance has been implicated in the pathogenesis of diabetes and insulin resistance [11]. Leptin also has an important inflammatory role responsible for endothelial dysfunction, increased oxidative stress, vascular inflammation and proliferation of vascular smooth muscle cells (VSMC) and resultant intimal hyperplasia [12].

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Vascular inflammation is the core pathological mechanism of diabetic microvascular complications and there is evidence that adipocytokines play a probable role in vascular inflammation and endothelial dysfunction [13].

A well-established relationship between leptin and insulin resistance, diabetes, obesity, inflammation and metabolic syndrome was proved in previous studies [14–16], but its relation to microvascular complications is still unclear. Moreover, microvascular complications are major health problems in Egypt [17]. This invited us to conduct the present study.

Materials and methods

This cross-sectional study was conducted among 120 participants with T2DM recruited from patients attending diabetes outpatient clinic of Alexandria Main University Hospital, Alexandria, Egypt who accepted to participate in the research after explaining the research aim.

Inclusion criteria included patients with T2DM and BMI > 18.5 kg/m^2 while exclusion criteria included ischemic cardiovascular event in previous 3 months, severe liver impairment, recent history of major trauma or surgery, hematological disorders or malignancy, chronic inflammatory or autoimmune diseases, as well as patients with recent history of sever significant infection at study entry.

This work was done in accordance with the Ethical Principles for Medical Research Involving Human Subjects outlined in the Helsinki Declaration in 1975 (revised in 2008). An approval was obtained from ethics committee of Faculty of Medicine, Alexandria University. All participates gave their written informed consent after explaining the nature and the aim of the study.

Participants were subjected to:

— Full medical history included following issues: personal data, detailed analysis of different cardio-metabolic risk factors, hypertension and dyslipidemia, history of macrovascular complications of diabetes (cardiovascular disease, cerebrovascular disease or peripheral arterial disease) and the use of antidiabetic, antihypertensive and antidyslipidemic drugs.

— Complete physical examination including body weight and height, body mass index (BMI) was calculated as body weight (kg) divided by body height squared (m^2). Waist circumference (WC) was measured at the midpoint between highest point of the iliac crest and lowest point of the costal margin at the end of normal expiration according to the WHO recommendations [18].

— Vital signs including pulse and blood pressure (BP) were measured. Neurological examination was performed and diagnosis of peripheral neuropathy was done based on abnormal results of more than one diagnostic test of the following: vibration perception threshold using a 128-Hz tuning fork, temperature perception, pin-prick, ankle reflex and touch-pressure sensation (10-g Semmes-Weinstein monofilament) [19].

— Fundus examination was done in ophthalmology outpatient clinic of Alexandria Main University Hospital using slit lamp biomicroscope plus fundus lens by expert ophthalmologist. Diabetic retinopathy was diagnosed based on fundus examination findings and was divided into proliferative and non-proliferative stages:
  - non-proliferative diabetic retinopathy (NPDR): presence of microaneurysms, hemorrhages and hard exudates;
  - proliferative diabetic retinopathy (PDR): presence of neovascularization.

Laboratory investigations

Blood was drawn for metabolic, biochemical and hematological parameters after a 12 hours overnight fasting and serum was used for measurement of the following: fasting plasma glucose (FPG) and fasting serum insulin [20]. Homeostasis Model Assessment 2 (HOMA2) calculator was used to estimate insulin resistance (%S) (HOMA-IR) according to the updated computer based HOMA2 mode. Whole blood was mixed in EDTA tubes for glycated heamoglobin (HbA_{1c}) [20]. Lipid profile was measured including total serum cholesterol, high density lipoprotein cholesterol (HDL-C), low density lipoprotein cholesterol (LDL-C) and serum triglycerides [20]. Serum was collected in Eppendorf tubes and kept at –80C till assay of leptin by using ELISA according to the manufacturer [21]. Urinary albumin/creatinine ratio (UACR) was measured in a random spot urine collection [22].

Serum creatinine was measured with calculation of eGFR for staging of diabetic kidney disease using CKD-EPI equation [23]. Diabetic kidney disease was diagnosed based on the presence of albuminuria (UACR ≥ 30 mg/g) and/or reduced eGFR (< 60 ml/min/1.73 m^2).

Statistical analysis of the data was done using IBM SPSS software package version 20.0. (Armonk, NY: IBM Corp). Qualitative data were described using number and percentage. The Kolmogorov-Smirnov test was used to verify the normality of distribution. Quantitative data were described using mean and standard deviation. Significance of the obtained results was judged at the 5% level.

Table 1. Characteristics of participants (n = 120)

Measures	Mean ± SD	
Age (years)	53.03 ± 7.65	
BMI [kg/m^2]	32.11 ± 3.26	
Waist circumference [cm]	110.4 ± 10.21	
FPG [mg/dL]	139.4 ± 46.88	
Insulin level [μIU/mL]	13.37 ± 10.67	
HOMA-IR	5.26 ± 3.26	
HbA_{1c} (%)	7.75 ± 1.66	
Total cholesterol [mg/dL]	212.7 ±49.79	
Triglycerides [mg/dL]	175.0 ± 77.59	
LDL-C [mg/dL]	107.7 ± 33.65	
HDL-C [mg/dL]	56.20 ± 6.44	
eGFR [ml/min/1.73 m^2]	85.40 ± 11.26	
UACR [mg/g]	52.08 ± 35.62	
Leptin [ng/ml]	20.26 ± 7.74	
Sex	No.	%
Male	58	48.3
Female	62	51.7
Duration of diabetes		
< 5 years	48	40.0
Peripheral neuropathy	26	21.7
Diabetic retinopathy	28	23.3
Proliferative	10	8.3
Non proliferative	18	15.0
UACR ≥ 30	34	28.3

CAD — coronary artery disease; BMI — body mass index; FPG — fasting plasma glucose; HbA_{1c} — hemoglobin A_{1c}; LDL-C — low density lipoprotein-cholesterol; HDL-C — high density lipoprotein-cholesterol; eGFR — estimated glomerular filtration rate; UACR — urinary albumin to creatinin ratio

The used tests were as follows:

— Mann-Whitney test: for abnormally distributed quantitative variables, to compare between two studied groups;

— Kruskal-Wallis test: for abnormally distributed quantitative variables, to compare between more than two studied groups;

— Spearman correlation coefficient was used to identify the correlation between the level of serum leptin and the other parameters in the studied subjects.

Statistical significance was set at P value ≤ 0.05.

Results

The present study was conducted among 120 participants with T2DM recruited from diabetes outpatient clinic of Alexandria Main University Hospital, Alexandria, Egypt. Participants' characteristics are mentioned in Table 1.

Table 2. Correlation between the level of serum leptin and the other parameters in the studied subjects

	Patients	
	r_s	P
Age (years)	0.167	0.068
Duration of DM (years)	0.280	0.002*
BMI [kg/m^2]	0.389	< 0.001*
Waist circumference [cm]	0.413	<0.001*
Systolic blood pressure [mm Hg]	0.383	< 0.001*
Diastolic blood pressure [mm Hg]	0.181	0.048*
FPG [mg/dL]	0.428	< 0.001*
Insulin [μIU/mL]	0.247	0.006*
HOMA-IR	0.323	< 0.001*
HbA_{1c} (%)	0.282	0.002*
Total cholesterol [mg/dL]	0.363	< 0.001*
Triglycerides [mg/dL]	0.338	< 0.001*
LDL-C [mg/dL]	0.331	< 0.001*
HDL-C [mg/dL]	0.086	0.350
eGFR [ml/min/1.73 m^2]	–0.273	0.003*
UACR [mg/g]	0.469	< 0.001*

r_s — Spearman coefficient; * — statistically significant at P ≤ 0.05
BMI — body mass index; FPG — fasting plasma glucose; HbA_{1c} — hemoglobin A_{1c}; LDL-C — low density lipoprotein-cholesterol; HDL-C — high density lipoprotein-cholesterol; eGFR — estimated glomerular filtration rate; UACR — urinary albumin to creatinin ratio

Serum leptin level was significantly positively correlated with diabetes duration, BMI, WC, systolic and diastolic blood pressure, FPG, HbA_{1c}, serum insulin level, HOMA-IR, total cholesterol, triglycerides and LDL-C (Table 2).

Correlation between serum leptin level and microvascular complications of diabetes

There was highly statistically significant positive correlation between serum leptin and UACR (Table 2, Figure 1), peripheral neuropathy and retinopathy (Table 3) (P value < 0.001) and statistically significant negative correlation between serum leptin and eGFR (Table 2, Figure 2) (P value < 0.001).

Discussion

Type 2 diabetes is a major health problem due to its high prevalence and the burden of its chronic complications [1]. Leptin is one of the adipokines incriminated in the pathogenesis of insulin resistance and T2DM. The present work was conducted to study the relation between serum leptin level and diabetic microvascular complications in a cohort of 120 participants with T2DM in Alexandria, Egypt. In the current study, serum leptin level was significantly positively correlated with

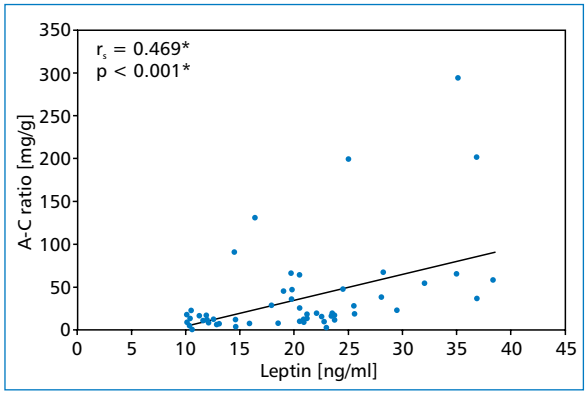

Figure 1. Correlation between leptin and UACR in studied subjects

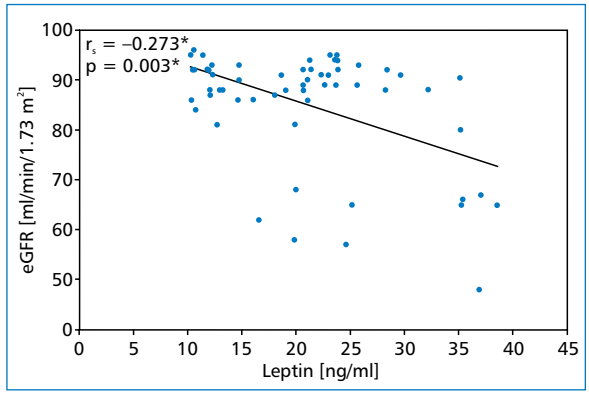

Figure 2. Correlation between leptin and eGFR in studied subjects

Table 3. Correlations between serum leptin level and peripheral neuropathy and retinopathy in the studied group

	N	Leptin Mean ± SD	Test of sig.	P
Peripheral neuropathy				
No	94	18.10 ± 6.14	U = 466.0*	< 0.001*
Yes	26	28.05 ± 8.01		
Fundus examination				
Normal	92	18.14 ± 6.20	H = 21.322*	< 0.001*
Proliferafive	10	29.26 ± 7.13		
Non proliferafive	18	26.09 ± 8.86		

U — Mann-Whitney test; H — H for Kruskal-Wallis test; p — p value for association between leptin and different parameters; * — statistically significant at P ≤ 0.05

diabetes duration, BMI, WC, systolic and diastolic blood pressure, FPG, HbA_{1c}, serum insulin level, HOMA-IR, total cholesterol, triglycerides and LDL-C. In agreement with the results of the present study, Cha et al. [24] found that plasma leptin level is positively correlated with BMI, fasting plasma glucose HbA_{1c} and total cholesterol values in patients with T2DM. Yassin et al. [25], who studied serum leptin level in patients with T2DM also found significant positive correlations between serum leptin and diabetes duration, cholesterol, triglycerides and LDL-C. Zulfania et al. [26] in concordance with the results of the present study found that serum leptin concentration was significantly correlated with BMI, FPG and HbA_{1c} in patients with T2DM.

Regarding the relation between serum leptin level and microvascular complications, the present study showed a significant positive correlation between serum leptin level and UACR, peripheral neuropathy and retinopathy and a significant negative correlation between serum leptin level and e-GFR. In concordance with the present study a meta-analysis by Rodríguez et al. [27] concluded that higher leptin levels were associated with microalbuminuria, macroalbuminuria and neuropathy, but in disagreement with the results of the present study, no association was found between serum leptin level and retinopathy. This difference may arise from the different patients' characteristics in studied cohorts. Cha et al. [24] showed similar results as they found a significant positive correlation between serum leptin level and urinary albumin excretion, and significant negative correlation with creatinine clearance. Yassin et al. [25] also showed a significant positive relation between serum leptin level and urinary albumin excretion and they concluded that serum leptin may be used as a marker of progression of diabetic kidney disease. On the other hand, Sari et al. [28], who studied the relation between serum leptin level and diabetic complications in patients with T2DM, found no significant difference between patients with and without diabetic nephropathy, retinopathy or neuropathy. Jung et al. [29], in concordance with the results of the present study, showed that serum leptin was significantly higher in patients with neuropathy than in patients without neuropathy. Uckaya et al. [30] studied the relation between serum leptin level and diabetic retinopathy and their results were similar to the

results of the present study as they found a significantly higher leptin level in patients with proliferative and non-proliferative diabetic retinopathy than in patients without retinopathy.

Conclusions

From the results of the present study we concluded that serum leptin level is associated with microvascular complications in T2DM. This may be due to the pro-inflammatory potential of leptin and its involvement in subclinical inflammation and endothelial injury which are incriminated in the pathogenesis of microvascular complications. Further studies may declare the link between serum leptin level and progression of microvascular complications and the use of serum leptin level as a prognostic factor in patients with microvascular complications.

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Conflict of interest

The authors declare to have no conflict of interest.

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Knowledge about diabetes mellitus among Polish medical students

ABSTRACT

Background. Due to high prevalence of diabetes mellitus all over the world it is essential for students who will become doctors of different specialties to possess a basic knowledge of it in this field. This pilot survey-based study was designed to assess diabetes related knowledge among faculty of medicine students from all of the medical universities in Poland.

Materials and methods. Students were invited to fulfill the questionnaire during Students' Diabetology Conference and via the Internet (social media). The survey consisted of questions about respondent's age and personal history of diabetes and diabetes related knowledge (etiology, symptoms, risk factors, complications and treatment of diabetes mellitus and additionally a section concerning gestational diabetes).

Results. A total number of 1200 medical students from Poland (70% women; mean age [SD] 22.12 [1.83] years of age) completed the survey. Mean test result was 66.62%. The best score was observed in the group of students enrolled in the 5th and 6th year of study, and those with diabetes mellitus type 1.

Conclusions. The study outcome proves that knowledge about diabetes mellitus among surveyed medical students in Poland is insufficient, therefore persistent improvement in transmitting it during the course of medical education is essential. (Clin Diabetol 2020; 9; 4: 245–252)

Key words: knowledge, diabetes mellitus, medical students, education, survey

Introduction

Among the most challenging health problems of the XXI century diabetes mellitus is one of the leaders [1]. It is estimated that 425 million adults all over the world are suffering from diabetes, whereas every second patient is undiagnosed [1]. According to the International Diabetes Federation (IDF), 629 million people in the world will have diabetes until the year 2045 [1]. In the Polish society, approximately 2.6 million people (i.e. almost 7% of the Polish population) are suffering from diabetes and 40% of patients are still undiagnosed [2]. The disease, especially when poorly controlled, may lead to many micro- and macrovascular complications. Diabetes doubles the risk of a cardiovascular disease and patients are prone to microvascular complications such as retinopathy, nephropathy and neuropathy what implicates the necessity for a patient to be treated by doctors of many different specialties. It is important to notice that diseases of different organs, (e.g. coronary artery disease, stroke, urogenital infections, foot ulcer) may be the first symptom of undiagnosed diabetes and doctors should advice the patient to have the blood glucose concentration tested [3].

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In the light of constantly increasing number of patients suffering from diabetes who visit many different health specialist during their lifespan, it is essential to propagate the broadest knowledge of the risk factors, symptoms and complications as well as treatment methods of the disease among medical students [1, 4, 5]. Being up to date with the guidelines and an ability to use obtained knowledge in practice is essential to ensure proper diabetes care [2]. An appropriate attention should be paid particularly to education in the field of risk factors and symptoms of diabetes mellitus in order to diagnose the disease early enough to prevent or delay its complications [6, 7]. During the past 10 years, there were only several studies performed in countries like Pakistan, India, Libya, Nepal, Saudi Arabia, Nigeria, Switzerland, Great Britain, Germany, United Stated of America and Poland which investigated medical students' knowledge related to diabetes [4–12]. These surveys included from 60 to approximately 400 respondents but to our knowledge, neither one evaluated diabetes related knowledge among students from the first to the last year of medical training, coming from all of the academic centers in one country.

The 6-year course of medical education in Poland is divided into a "preclinical" period (year 1th and 2nd) and a "clinical" one (years 3rd–6th). Knowledge about pathophysiology of the disease is obtained during a preclinical phase, whereas information about treatment, complications and prognosis is gained in further years of education. Knowledge gained during medical studies is crucial for young doctors and in some medical specialties university education is the only time in once career to learn about diabetes mellitus [12]. The aim of this study was to assess diabetes related knowledge among faculty of medicine students from all of the medical universities in Poland.

Material and methods

This was a survey based, multi-centered study conducted among Polish medical faculty students from all medical universities in Poland during the Diabetology Conference in Zabrze on 21st of November 2015 organized by the International Medical Federation of Students' Association IFMSA-Poland. After the Conference ended the survey assessing diabetes related knowledge was shared among faculty of medicine students studying in all of medical universities in Poland by social media as this form was the best possible way to reach respondents from all of the country. Each medical university representative student attending the Conference was asked to carry on the survey to their fellow students in the representative's university throughout the Internet with the use of Google Forms till the 6th of December 2015. The questionnaire was self-prepared and approved by two independent experts in diabetology field and was pre-tested on a group of 60 randomly selected students of the Medical University of Silesia in Katowice. Underlying components were identified using principal component analysis and the internal consistency of questions was checked. The questionnaire consisted of 19 questions, including 13 single-choice questions and 6 multiple-choice questions and was divided into the following categories: etiology, symptoms, risk factors, complications and treatment of diabetes mellitus and a section concerning gestational diabetes (Table 1). Evaluation process of the survey was performed in the following manner: a single-choice question was awarded 1 point for a correct answer, 0 points for a wrong one or the answer "I do not know"; each multiple-choice question was awarded 1 point for each correctly selected answer and minus 1 point for a wrong answer indicated. One could score up to 33 points in the pool. Students were also asked to indicate their age, a personal history of diabetes mellitus and the year of studies. Participants were split into two groups depending on the year of study ("preclinical group" which is 1st–2nd year of faculty of medicine and "clinical group" which is 3rd–6th year of faculty of medicine).

The results obtained were analyzed using STATISTICA 12.5 (StatSoft, Cracow, Poland). Quantitative variables were compared using Mann-Whitney U test. Kruskal-Wallis test was used to compare the multiple groups and the Spearman's rank correlation to verify the relationship between variables. A P value of < 0.05 was considered significant.

Results

1200 (70% women; mean age [SD] 22.12 [1.83] years of age) students from all of the medical universities in Poland completed our diabetic knowledge test, which is approximately several percent of all the medical students in Poland. 1.7% (n = 20) of the respondents declared to have type 1 diabetes mellitus and none declared to have type 2 of the disease. Mean result obtained from the test was 66.62%, whilst the worst one was 29.4% and the best was 100%. The distribution of the results in the study cohort are presented according to the year of study (Figure 1). There was no significant difference in the test result in relation to gender (66.53 ± 11.6% *vs.* 66.64 ± 11.12%; $P > 0.05$) in men and women respectively. The higher was the student's year of study, the better was the test result score (Figure 1). Students who started or continued clinical subjects achieved significantly higher result in comparison with "preclinical" students' group

Table 1. Diabetes related knowledge questionnaire (correct answers are in italics)

1. Is diabetes mellitus (DM) an infectious disease?
   a) Yes
   b) No
   c) I do not know
2. Insulin:
   a) Increases blood glucose concentration
   b) Decreases blood glucose concentration
   c) Does not affect the blood glucose concentration
   d) I do not know how it affects blood glucose concentration
   e) I do not know what insulin is
3. Too low blood glucose concentration:
   a) It does not affect one's health
   b) May be dangerous for life
   c) Causes cancer
   d) It is beneficial in DM because it makes it easier to lose unnecessary kilograms
   e) I do not know
4. What is a proper fasting plasma glucose concentration?
   a) Less than 70 mg/dl
   b) 70–99 mg/dl
   c) 100–125 mg/dl
   d) Above 125 mg/dl
   e) I do not know
5. What are the risk factors of DM type 2? More than 1 answer is correct:
   a) Inadequate diet
   b) Smoking cigarettes
   c) Sedentary lifestyle
   d) Alcohol consumption
   e) Hypertension
   f) Genetic predisposition
   g) Obesity
   h) Frequent use of antibiotics
   i) I do not know
6. What may be the symptoms of DM? More than 1 answer is correct:
   a) Polyuria
   b) Oliguria
   c) Polydipsia
   d) Oligodipsia
   e) Increased activity
   f) Somnolence
   g) Increased appetite
   h) Blurred vision
   i) Muscle pain
   j) Unexpected weight loss
   k) I do not know
7. What can untreated or poorly controlled diabetes lead to? More than 1 answer is correct:
   a) Diabetic nephropathy
   b) Sepsis
   c) Coronary artery disease
   d) Vascular damage
   e) Osteoporosis
   f) Alzheimer's disease

→

Table 1 (cont.). Diabetes related knowledge questionnaire (correct answers are in italics)

g) Blindness
h) Stomach cancer
i) Diabetic foot
j) I do not know

8. Is it possible to die from complications of DM?
a) Yes
b) No
c) Yes, but very rarely
d) I do not know

9. Is correct blood glucose concentration important for pregnant women?
a) Yes
b) No
c) I do not know

10. Should pregnant women be screened for DM?
a) Yes
b) No
c) I do not know

11. What is the basic treatment of DM besides medications?
a) Medications only
b) Physical effort
c) Proper diet
d) B and C
e) I do not know

12. What can be used to treat DM?
a) Insulin
b) Oral medications
c) Diet
d) All listed
e) I do not know

13. What is the first choice treatment in DM type 1?
a) Metformin
b) Sulphonylurea
c) Insulin
d) Acarbose
e) DPP-4 inhibitors
f) I do not know

14. Apart from dietary intervention, how can gestational diabetes mellitus (GDM) be treated?
a) Sulphonylurea
b) Acarbose
c) Gliflozins
d) Insulin
e) GLP-1 receptor agonists
f) I do not know

15. Glycated hemoglobin (HbA_{1c}) determines the average blood glucose concentration during past:
a) Week
b) 2 weeks
c) 1 month
d) 3 months
e) Half a year
f) I do not know

→

Table 1 (cont.). Diabetes related knowledge questionnaire (correct answers are in italics)

16. The criterion for good controlled DM type 2 for general population is the HbA_{1c} level:
    a) ≤ 6.0%
    b) ≤ 6.5%
    c) ≤ 7%
    d) ≤ 8.0%
    e) ≤ 10.0%
    f) I do not know
17. How can insulin be administered? More than 1 answer is correct
    a) Intradermally
    b) Subcutaneously
    c) Intramuscularly
    d) Intravenously
    e) Orally
    f) I do not know
18. Which insulin could be administrated intravenously? More than 1 answer is correct
    a) Lispro
    b) Neutral insulin
    c) NPH
    d) Glargine
    e) Insulin cannot be administered intravenously
    f) I do not know
19. What are the differences between human and analogue insulin?
    a) Human insulin can be injected just before a meal, analog insulin should be injected 30 minutes before a meal
    b) Human insulin is injected just like an analogue insulin after the meal
    c) Human insulin should be injected half an hour before a meal and analogue insulin can be injected just before a meal
    d) There is no need to eat snacks with human insulin
    e) I do not know

DM — diabetes mellitus; GDM — gestational diabetes mellitus; HbA_{1c} — glycated hemoglobin

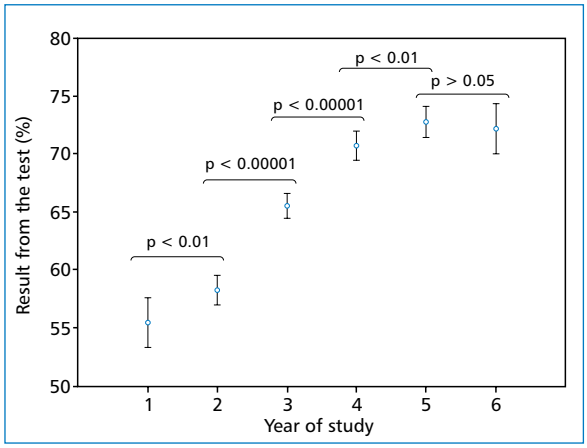

Figure 1. The distribution of the results in the study cohort according to the year of study

(70 ± 9.85 *vs*. 57.56 ± 9.59; P = 0.0000) (Table 2). As a conclusion, being in the group of clinical education, higher year of study and having type 1 diabetes mellitus (P = 0.0084) were the factors significantly associated with better results of the diabetes knowledge test.

Discussion

According to our best knowledge, this investigation is the biggest multi-centered study not only in Poland, but also worldwide, since we have obtained 1200 respondents studying medicine faculty from the 1st to the 6th year of study from all of the medical universities in Poland. As patients suffering from diabetes mellitus are prone to many complications which are treated by doctors of different specialties, the basic knowledge of diabetes mellitus is crucial for every doctor. It might happen that before a patient visits a diabetologist, they are first seen by a general physician, surgeon, urologists, ophthalmologist, cardiologist, nephrologist or neurologist who should be able to diagnose the disease properly based on declared symptoms. Taking into consideration that the prevalence of diabetes is rising, medical students should be well-educated about the disease during their course of studies.

Table 2. Percentage of students answering correctly about risk factors, symptoms and complications of diabetes with regard to preclinical and clinical level of education

Variable	Preclinical students	Clinical students	P value
Risk factors			
Unhealthy diet	96% (n = 294)	96% (n = 792)	0.9522
Sedentary lifestyle	89% (n = 273)	95% (n = 777)	0.0040
Hypertension	32% (n = 97)	36% (n = 296)	0.1897
Genetic predisposition	76% (n = 233)	89% (n = 731)	0.0000
Obesity	95% (n = 292)	99% (n = 815)	0.0005
Symptoms of diabetes			
Polyuria	74% (n = 229)	97% (n = 793)	0.0000
Polydipsia	89% (n = 273)	99% (n = 812)	0.0000
Somnolence	91% (n = 277)	94% (n = 776)	0.0369
Blurred vision	56% (n = 174)	73% (n = 600)	0.0000
Unexpected weight loss	41% (n = 125)	63% (n = 518)	0.0000
Complications of diabetes			
Diabetic nephropathy	87% (n = 265)	98% (n = 799)	0.0000
Coronary artery disease	54% (n = 166)	69% (n = 566)	0.0000
Vascular damage	73% (n = 224)	96% (n = 784)	0.0000
Blindness	85% (n = 260)	98% (n = 801)	0.0000
Diabetic foot	97% (n = 297)	99% (n = 815)	0.0444

n — number. A P value of < 0.05 is considered statistically significant

Studies related to medicine faculty students' knowledge about diabetes performed up to date are scarce and it is impossible to directly compare the outcomes because of different groups of respondents and different surveys applied. For this reason we will only refer to them to present just a general insight into studies dealing with assessment of medical faculty students' awareness of diabetes. The general level of diabetes related knowledge assessed in the presented research is unsatisfying. The lowest score was achieved by a 1st year medical students, which reflects the fact that people entering universities have very little knowledge of diabetes. Only few students achieved 100% score, and one might suspect that it could be due to an individual interest in diabetology or having to deal on everyday basis with a relative having diabetes. Students have essential problems with marking correctly all risk factors, symptoms and type of insulin applied intravenously. What is worth emphasizing, only 50% of respondents knew what medications could be administered to pregnant woman suffering from diabetes. Sagar et al., in a study conducted in Libya testing the knowledge of 325 final year medical students using an Arabic 24-item "Short Diabetes Knowledge Test", assessed students' knowledge as reasonable (mean score 76.7%), with major deficiencies in dietary management [8]. Our respondents demonstrated knowledge on a lower average level (mean score 66.6%), however, it is impossible to compare them directly as the surveys differed. Yet the general author's conclusion was that students in Libya have a proper working knowledge of diabetes and would be able to care for patients [8]. An opposite observation was made by Lansang et al. from the University of Florida in United States of America, who noticed that medical students are unable to translate theoretical knowledge into practice [13]. Students in that study were able to diagnose diabetes but they could not indicate appropriate scheme of treatment. Students who took part in our research recognized symptoms of hyperglycemia properly, however our respondents had significant problems with questions related to insulin use, which was also observed in the American research indicated above. In the context of a high number of people with hypertension, medical student should be aware that it is a risk factor for diabetes [14], the more so that hypertension was poorly recognized as a risk factor among entire study group of our students. Students' knowledge should be at least satisfactory regarding symptoms of diabetes mellitus what is essential for patients prompt diagnosis which nowadays is still delayed. Both preclinical and clinical students associated diabetes with somnolence (91% of preclinical students *vs.* 95% of clinical ones) and polydipsia (89% *vs.* 99%). The least known symptom of diabetes in both groups was an unexpected weight loss (41% *vs*. 63%) and visual impairment (56% *vs.* 73%).

Our findings are in accordance with other researchers', one of whom is Mumtaz et al., in Pakistan, as the most frequently marked symptoms in their study were also polyuria and polydipsia among preclinical and clinical students (81% *vs*. 90% respectively) while weight loss was associated with diabetes by 39% of preclinical students and by 57% clinical students, which is also comparable to our results [4]. As a conclusion, in different countries both polyuria and polydipsia are recognized by majority of students as a potential symptom of diabetes, however, sign such as unexpected weight loss is overlooked. In the field of identifying complications, pointing coronary disease as one of them, was problematic for both clinical and preclinical groups of students. Only 68% of clinical students and 53% of preclinical students responded correctly to that question ($P < 0.001$). In the Pakistani study similar results were obtained with 72% of the clinical and 51% of the preclinical students being able to indicate the diabetes complications correctly [4]. Among our respondents 1.7% declared to suffer from type 1 diabetes mellitus and they obtained a better result from the test compared to healthy colleagues (73.4% *vs*. 66.5%), which is obviously explained by the fact that people having diabetes are supposed to be educated while treatment. A study conducted in Saudi Arabia showed a significant correlation between gender and the level of knowledge about diabetes, where men have obtained a better result [9]. In our study there was no significant difference between the level of knowledge among men and women. It is worth mentioning that the gap in knowledge related to pointing hypertension as a risk factor of diabetes and weight loss as a symptom of the disease is the same as in general population what we have proven in our previous research [15]. Taking into consideration all students attending medicine course in Poland, our respond rate is estimated to be a couple of percent, which is the main limitation of our study. It is difficult to preciously assess the number of students in medical faculty each year because not every university publishes such data. On the other hand, it is just a pilot study and it already has the biggest total number of medicine faculty students in Poland and worldwide and the poor results obtained enquire to continuation of research in this field. The low response rate is a well-recognized limitation of Web-based survey [16], which gives the possibility to reach many different parts of the country as well as bigger number of people that paper surveys, however not every respondent expresses a desire to take part in this kind of research. Similar observations related to low response rate to invitation related to participation in the questionnaire based study come from our previous research related to diabetes knowledge assessment where we examined Polish mountain guides [17] and people from the general community [15].

Conclusions

It the light of constantly increasing prevalence of diabetes mellitus which affects many organs, medical faculty students, who will became doctors of different specialties should gain a basic diabetes related knowledge during their university studies. Barring in mind the limitation of the ability to reach the high response rate, the presented study indicates that there are gaps in students' knowledge related to diabetes what needs a future attention and indicates a need for persistent improvement in spreading diabetology knowledge during medical education in order to struggle more efficiently against diabetes epidemic.

Conflict of interest

The authors declare to have no conflict of interest.

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Bolus calculator in personal insulin pumps — advantages, differences and practical tips

ABSTRACT

The widespread use of personal insulin pumps in the treatment of type 1 diabetes has significantly improved the effects of therapy. Better metabolic control of diabetes, but also increased comfort of life and professional opportunities have been achieved. One of the functions that increased the effectiveness of insulin therapy using a personal insulin pump is the bolus calculator. The bolus calculator function allows the user to dose their insulin more accurately before meals and to correct hyperglycaemia. The bolus calculator algorithms in insulin pumps of individual manufacturers differ in the way they calculate the correction dose and the amount of so-called active insulin. Consequently, with the same treatment parameters, individual bolus calculators may offer a different dose of insulin. Understanding the principles of the bolus calculator by therapeutic team members and patients is very important for proper diabetes education and diabetes management. (Clin Diabetol 2020; 9; 4: 253–258)

Key words: type 1 diabetes, personal insulin pump, bolus calculator, active insulin

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Introduction

People suffering from type 1 diabetes should be treated with intensive functional insulin therapy from the very beginning. This approach is based on the administration of variable doses of short-acting insulin (bolus insulin) and long-acting insulin (basal insulin). This method of treatment can be implemented using pen injectors or personal insulin pump (PIP). Precise adjustment of insulin doses to the daily activities of patients is much easier with the use of PIP, because continuous subcutaneous insulin infusion (CSII) allows for better mimicking of the physiological secretion of insulin. This method of insulin administration improves metabolic control of diabetes, reduces the incidence of hypoglycaemia and improves the quality of life of patients with type 1 diabetes [1–4]. One of the most important features of PIP is a bolus calculator (BC). It is recommended to use BC from the beginning of therapy [5]. This facilitates the precise estimation of insulin bolus [6, 7]. In addition, BC provides very important information about the amount of active insulin in the body, so-called insulin-on-board (IOB). This parameter takes into account insulin boluses delivered within the last few hours and prevents administration of a too high correction dose [8]. Studies have also shown that the use of BC improves glycaemic control in patients by reducing blood glucose variability, i.e. lowering the low blood glucose index (LBGI) and high blood glucose index (HBGI) parameters [9]. Benefits of using PIP and BC are possible in well-educated patients who understand the principles of its function [10].

Bolus calculator in various models of insulin pumps

Bolus calculators can be integrated in insulin pumps, stand-alone devices or software applications:

— bolus calculator integrated in PIP (Medtronic): Minimed 715, Minimed 722, Minimed VEO, Minimed 640G [11];
— external bolus calculator — a remote control that functions as a blood glucose meter (Roche): Accu-Chek Spirit Combo, Accu-Chek Solo [12];
— bolus calculator in the mylife™ App (Ypsomed) that can be installed on a mobile device, e.g. a smartphone: YpsoPump — one-way communication between PIP and the BC application; data from PIP are automatically sent to the mobile device; bolus administration via the application is not possible [13].

Basic parameters common to all bolus calculators

The basic element of intensive functional insulin therapy is the systematic use of a well-programmed bolus calculator, taking into account the individual needs of the patient. The parameters that the bolus calculator takes into account include [14]:

— **insulin-to-carbohydrate ratio (ICR)**. As in the case of injection therapy, it is necessary to determine ICRs for each time interval during the day, i.e. what dose of insulin will cover the consumption of 1 carbohydrate exchange (1 CE = 10 g of carbohydrates) or how many grams of carbohydrates compensates for 1 unit of insulin. The user can select how to enter carbohydrates into the BC, with the exception of the BC on the YpsoPump, which can only be programmed in grams, not KE. Bolus calculators give the users greater flexibility in establishing insulin-to-carbohydrate ratios and precise insulin dosing with accuracy of 0.1 unit. Accuracy of less than 0.1 unit may be achieved with some pump models, but this is not usually the case in clinical practice.
Examples of programming the insulin-to-carbohydrate ratios:
1 unit/CE or 10 g of carbohydrates per 1 unit of insulin,
1.4 units/CE or 7 g of carbohydrates per 1 unit of insulin;
— **insulin sensitivity (correction factor)** — this parameter determines how much blood glucose will decrease (in mg/dL) after administration of 1 unit of insulin. For most adult patients the values of this parameter are in the range of 30–60 mg/dL and may be different in particular times of the day;
— **target blood glucose** — this is the desired blood glucose value at the time of bolus administration. Depending on the BC, it is possible to set a single blood glucose value or target range, e.g. 80––120 mg/dL. The settings determine the amount of the correction bolus and the so-called negative correction, i.e. reduction of the meal bolus in the event of blood glucose levels below the target value. Target blood glucose should be based on clinical status, current metabolic control of diabetes, and patient expectations (Fig. 1). This parameter should not be confused with the postprandial blood glucose target, which is not included in BC programming.

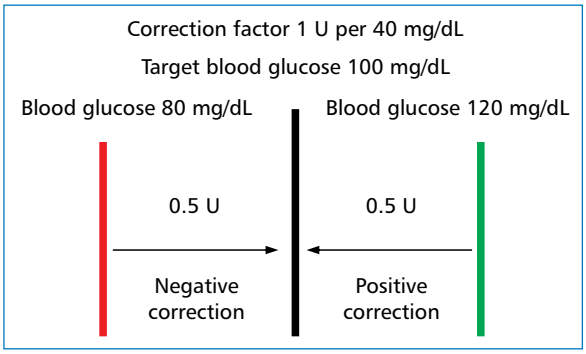

Figure 1. Calculation of a negative or positive correction by the bolus calculator for a sensitivity factor of 1 U per 40 mg/dL

Differences in programming target blood glucose depending on PIP model

Insulin pumps produced by Medtronic and Roche are programmable for either a single target blood glucose value or a target range. The difference between them is a different algorithm for calculating the correction bolus. Medtronic's BC uses the border values of the set range as glycaemic targets. For example, when target blood glucose of 80–120 mg/dL is programmed, then if a hypoglycaemia occurs, the BC will calculate a negative correction for the target blood glucose of 80 mg/dL, and in the case of hyperglycaemia, it will use 120 mg/dL as the target blood glucose. For Roche's BC, the correction bolus is always computed to the middle value of the range, i.e. with a blood glucose target of 80–120 mg/dL — for both hypo- and hyperglycaemia, it will choose 100 mg/dL as the desired value (Fig. 2). Users of the bolus calculator in YpsoPump can only set up a single blood glucose value; there is no option to program a wider range.

Active insulin time (insulin on board) — determines how long the BC algorithm will include bolus insulin. On this basis, PIP informs about the amount of the active insulin that is already in the body. This feature is designed to prevent too much insulin being delivered to correct hyperglycaemia while insulin from previous

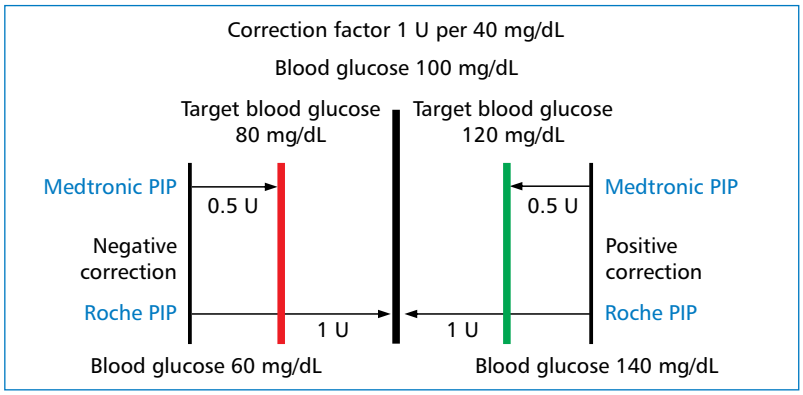

Figure 2. Calculations of the correction dose of insulin by the bolus calculator depending on the manufacturer; PIP — personal insulin pump

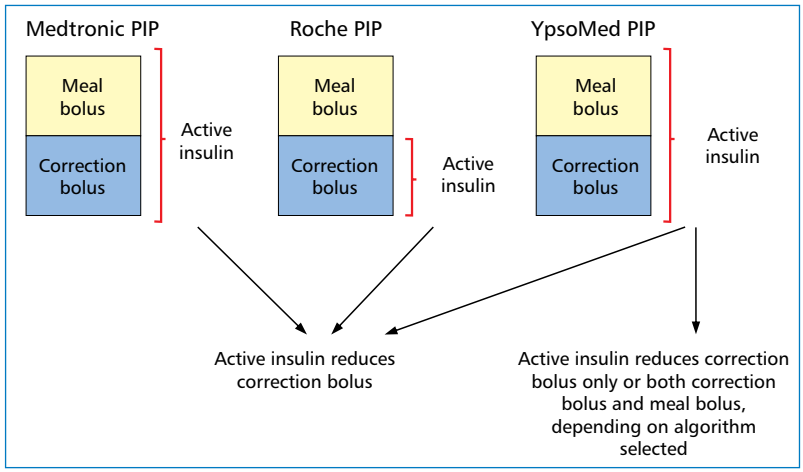

Figure 3. Calculating insulin acting time/insulin-on-board depending on the manufacturer; PIP — personal insulin pump

boluses is still working. Therefore, active insulin feature reduces only the dose of the correction bolus, not the meal bolus. An exception is one of the two available BC algorithms in the mylife™ App, where this feature reduces both the correction dose and mealtime bolus. The choice of algorithm in the mylife™ App depends on the patient's preferences and the recommendations of the diabetologist. In any bolus calculator, the amount of active insulin and the rate at which it is decreasing is directly related to the programmed insulin acting time. Information about the amount of active insulin is available, depending on the PIP model, from the pump screen, remote control or app.

The algorithm that calculates the amount of active insulin differs between manufacturers. The BC in PIPs by Medtronic and YpsoMed considers both mealtime and correction bolus insulin to be active insulin. In Roche's insulin pump, active insulin comes only from correction boluses (Fig. 3).

The BC algorithm in Accu-Chek Spirit Combo and Accu-Chek Solo takes into account additional parameters along with the insulin acting time, which include:

— **meal rise** — shows the maximum value by which blood glucose can rise after a meal without prompting a correction bolus;
— **offset time** — is period of time after a bolus administration until significant reduction in blood glucose begins. This is the first phase of the period known as insulin acting time;
— **snack size** — is the amount of carbohydrates that is not considered as a standard meal and the BC does not activate the meal rise, so that the blood glucose increase reading is recorded and prompts a correction bolus.

The manufacturer's educational materials explain the bolus calculator algorithm using a trapezoid diagram, where the longer base of the trapezoid is "insulin acting time", the shorter base is "offset time", and

the height of the trapezoid is the "meal rise' parameter. Increasing/decreasing these parameters causes a corresponding increase/decrease in the trapezoidal area, which in practice means a reduced/increased correction dose of insulin at the time of postprandial hyperglycaemia.

Clinical aspects of the bolus calculator

Programming the BC in accordance with the principles of intensive functional insulin therapy requires extensive experience of the doctor and good cooperation with the patient. Proper blood glucose self-monitoring using a blood glucose meter or continuous glucose monitoring is essential. The bolus calculator is especially appreciated by professionally active people because it saves time and improves postprandial blood glucose levels [15].

Typically when setting the BC, the following parameters must be determined individually for each patient:

— **insulin-to-carbohydrate ratio (ICR)** — to initially determine this parameter, the rule of 400 should be used. Dividing 400 by the total daily insulin dose (DDI) gives the number of grams covered by 1 U of insulin, e.g. 400: 40 U = 10 g/U. It is an average value for the whole day. Insulin sensitivity changes during the day, so ICR in the morning hours can be up to 50% higher than that calculated and it can be lower in the midday hours;

— **insulin sensitivity (correction factor)** — one recommended approach is the 1800 rule. Dividing 1800 by the DDI gives the average blood glucose reduction after administration of 1 U of insulin, e.g. 1800: 40 U = 45 mg/dL. Insulin sensitivity shows high individual and daily variability. The highest insulin sensitivity is observed in the first half of the night and in the middle of the day, and it is lower in the morning and during afternoon and evening hours;

— **target blood glucose** — it is preferable that the daytime values of this parameter should be lower than the nighttime values, because the perception of hypoglycaemia at night is impaired. In clinical practice, these values are in the range of 80–120 mg/dL. In some clinical situations, this parameter requires individual adjustment, e.g., in pregnant women with diabetes, the recommended target blood glucose at night and before meals is 70––90 mg/dL. Patients with known proliferative retinopathy and chronic hyperglycaemia require higher glycaemic targets, e.g., 120–150 mg/dL. Similarly, patients with hypoglycaemia unawareness or have fear of hypoglycaemia will need higher blood glucose levels;

— **active insulin time (insulin on board)** — in clinical practice, active insulin time/IOB in adults is programmed for 3–4 hours. It depends on the size of the bolus administered, and for boluses greater than 10–15 units, this time can be extended to 5 hours. During pregnancy, when postprandial glucose control is very important and frequent correction boluses are required, the insulin acting time is usually programmed for 3 hours. Some pump models offer the option of programming the insulin acting time with an accuracy of 15–30 minutes. Information about the amount of active insulin has a significant impact on the therapeutic decisions made by the patient [16]. A special situation is when hyperglycaemia persists for a long time and the patient administers correction boluses repeatedly. Active insulin function then reduces the risk of hypoglycaemia due to the simultaneous action of several boluses. In everyday life, decisions about the correction of hyperglycaemia just before going to bed are a frequent problem. The BC allows the user to calculate a safe dose of a correction bolus, and data on the amount of active insulin makes it easier to decide on the consumption of an additional portion of carbohydrates.

Bolus insulin must be taken into account during physical activity. The bolus calculator is a tool that allows the user to adjust the amount of active insulin to their training in a repeatable and precise manner [17]. The effect of physical activity is very individual and only on the basis of the patient's own experience it is possible to determine what amount of active insulin is appropriate for him or her. Too much active insulin requires the consumption of carbohydrates. This information is especially valuable when the patient plans to disconnect the insulin pump, as too little active insulin can cause a significant increase in blood glucose and lead to ketosis.

The specific features of BC in the Accu-Chek Spirit Combo System are presented in Table 1. The initial BC settings were proposed by a group of experts of the Diabetes Poland (PTD) (Table 1) [18]. These settings require clinical verification and are only a starting point for the use of BC. The same BC programming principles apply to the Accu-Chek Solo.

Correct use of a bolus calculator by patients

Optimal use of a BC requires the patient to enter information about the amount of carbohydrate consumed and the current blood glucose level. It is necessary to accurately count the carbohydrate consumed, which is entered into the calculator in grams

Table 1. Proposed bolus calculator settings in the Accu-Chek Combo system (modified after [18])

Parameter (available range)[1]	Units	Individual settings				
		1–6 years	7–11 years	12–18 years[2]	Adults	Women
Carbohydrate exchange (KE*)		KE	KE	KE	KE	KE
Insulin sensitivity, units/1 KE (0.1–24)	U/KE	The 400 rule	The 400 rule	The 400 rule	The 400 rule	The 400 rule
Insulin sensitivity, blood glucose reduction by 1 unit of insulin		The 1800 rule	The 1800 rule	The 1800 rule	The 1800 rule	The 1800 rule
Bolus advice options						
Meal rise (50–200)	mg/dL	80	80	50	50	50
Snack size (0–2.4 KE)	KE	0.5–1.0	1.0–1.5	2	2	2
Insulin acting time (1.5–8 hours)	h	3	3	3	3–5[3]	3–4[3]
Offset time (from 45 min to blood glucose reduction)	h	1.5	1.5	1	1	3/4–1[4]

[1]Definitions of bolus calculator features are presented in the respective operating manual
[2]Parameter settings can be applied as for adults
[3]Depending on bolus size, 3 h < 10 U, 4 h < 10–20 U, 5 h > 20 U
[4]Depending on the trimester of pregnancy
*Germ. Kohlenhydrateinheit

or carbohydrate exchanges. Blood glucose values are sent wirelessly from the glucose meters to the pump, or they can be entered manually. Only for the Accu-Chek Performa Combo remote control, which is also a glucometer, the blood glucose results cannot be entered manually. After the measurement, blood glucose is stored in the remote control for 5 minutes.

The insulin dose proposed by the BC on the basis of the above parameters will not always be adequate to the current clinical situation [19]. This also applies to the patient's decision to choose a combination or extended bolus type. A well-educated PIP user will not always agree with BC's suggestion. For example, when planning physical exercise within 2 hours of a meal, the patient should reduce the calculated bolus by 30–50% in order to avoid hypoglycaemia. Conversely, increasing the bolus dose will be required during periods of increased insulin requirements. These includes situations that are very common in everyday life, such as infections, limited physical activity, stress or eating meals with a high glycaemic index. Then, the standard BC settings do not match the actual insulin requirement. The bolus calculator also does not take into account the variable rate of insulin absorption, gastric emptying, or the qualitative composition of the meal.

It is worth noting that repeated non-compliance with the BC may indicate the need to modify its settings. Available software for the analysis of data from insulin pumps (Accu-Chek SmartPix Software, CarelinkPro, mylife Software) make it possible to assess what percentage of boluses calculated by BC is modified by the patient.

Another important issue with the use of BC is the dosing of insulin for meals rich in proteins and fats. In the bolus calculators available on the market, it is not possible to separately enter information about the number of consumed protein-fat exchanges. The insulin dose can be increased by entering additional CEs or modified immediately after the BC calculation [20].

Conclusions

The bolus calculator is an indispensable tool in the treatment of type 1 diabetes mellitus with continuous subcutaneous insulin infusion. There is a lot of evidence for improving the quality of life and metabolic control of diabetes. It is recommended to use a BC from the beginning of therapy with PIP. Diabetes education and close cooperation with a diabetologist is necessary so that the patient knows how the bolus calculator works.

Conflict of Interest

UF received a lecture fee from Ypsomed. AG and DZZ participated in the Advisory Board for Medtronic, Roche Diagnostics and Ypsomed and received a lecture fee from Medtronic, Roche Diagnostics, and Ypsomed. AlA has received lecture fees from Medtronic, Roche Diagnostics, and Ypsomed. MM and AnA have received lecture fees from Ypsomed.

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REVIEW ARTICLE
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Cinnamon supplementation improves blood pressure in type 2 diabetic patients: a systematic review and meta-analysis of randomized controlled trials

ABSTRACT

Some studies have suggested that consumption of some herbal medicines may improve blood pressure. The present systematic review and meta-analysis was conducted to assess the efficacy of cinnamon supplementation on blood pressure in type 2 diabetic patients.

The systematic search was undertaking using several online databases (PubMed, Embase, Scopus and Web of Sciences) to identify randomized controlled trials (RCTs) investigating the effect of cinnamon supplementation on systolic blood pressure (SBP) or diastolic blood pressure (DBP) in type 2 diabetic patients that were published up until 10 December 2019. Potential publication bias was assessed using the Egger regression test.

Five full-text articles were included in this meta-analysis. Pooled results of the meta-analysis on 332 participants indicated a significant reduction in SBP and DBP following cinnamon administration. No publication bias was found.

The results of the present study suggested that cinnamon might be effective in improving blood pressure in type 2 diabetic patients. (Clin Diabetol 2020; 9; 4: 259–266)

Key words: cinnamon, blood pressure, cardiovascular, nutrition, meta-analysis

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Introduction

Hypertension is one of the most common adult chronic disorders and affects approximately 25% of the adult population and 8.5% of diabetic patients globally. The complications of hypertension is also estimated to cause 7 million deaths annually [1, 2]. There is an urgent need to improve the control blood pressure in hypertensive patients, as guideline recommended targets are often not adequately attained using current drug treatment. It has been reported that only approximately 50% of people with hypertension have controlled BP (less than 140/90 mm Hg), whereas more than 13% of patients have systolic (SBP) > 160 mm Hg and/or diastolic blood pressure (DBP) > 100 mm Hg [3]. Moreover, the complications and adverse effects of antihypertensive drugs as well as their high costs appear to reduce the adherence to treatment of hypertensive patients [4]. Hence alternative, or complementary medicines, for hypertension are receiving some interest.

Recently, there has been more attention focused on using nutritional and bioactive compounds for prevention of chronic illnesses and enhancement of

well-being [5]. Cinnamomum (Cinnamon) is a genus from the Lauraceae family of plants and is considered to be a dietary herbal medicine [6]. Cinnamon has been reported to have numerous properties, including antidiabetic, anti-inflammatory, lipid-lowering, antioxidant, antimicrobial, and anticancer properties [7]. It has also been suggested that it could be used as an herbal remedy for the treatment of several disorders including cardiovascular diseases, type 2 diabetes, chronic digestive difficulties, and Alzheimer's disease [8]. Some previous studies have reported that cinnamon supplementation can significantly reduce systolic and diastolic blood pressure in the subjects with type 2 diabetes mellitus [9–11]. The results of some other studies have not been consistent [12, 13]. Therefore, the effect of cinnamon intake on the systolic and diastolic blood pressure remains unclear. Disagreements among the findings of published trials might be due to the differences in dosage of cinnamon used, study design, characteristics of study populations, and duration of the trials.

The present meta-analysis aimed to assess the effect of cinnamon supplementation on the blood pressure and anthropometric parameters as known critical risk factors for hypertension.

Methods

The present meta-analysis was conducted as recommended by the Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA) [14].

Systematic search

Our systematic search was undertaken using several online databases: PubMed, Embase, Scopus and Web of Sciences, to identify randomized controlled trials (RCTs) investigating the effects of cinnamon supplementation on blood pressure in type 2 diabetic patients for all potential publication from the earliest available time up to 10th December 2019. The following search strategy was used without any time or language limitation: [Keywords for supplement] AND [Keywords for outcomes] AND [Keywords for disease] AND [Keywords for study design]. To complete search process, Google scholar, Cochrane Library and list of relevant references were then hand-searched to identify eligible papers that might have been missed.

Inclusion and exclusion criteria

Relevant articles were selected for meta-analysis that met the following inclusion criteria: (1) being a RCT, (2) using cinnamon as supplement in confirmed type 2 diabetic patients, (3) reporting sufficient data for baseline and final trials of SBP or/and DBP in both cinnamon and control groups, (4) human studies and (5) English languages publication. Unpublished data and gray literature, such as conference abstracts, book chapters, editorials and letters and the like, studies conducted on other types of diabetes or related disorders, healthy and just glucose-intolerant subjects were excluded.

Data extraction

Eligible articles were abstracted by two reviewers separately and following information were extracted using a pre-defined form: last name of first author, publication year, country, sample size, age of participants, target population, intervention duration, dose of cinnamon, type of the supplement and mean changes/ /standard deviation (SD) of the outcomes. Net changes were calculated by subtracting pre-test from post-test. Also, SD for changes was obtained using the following formula: $\sqrt{([(SD\ pre)^2 + (SD\ post)^2] - [2r \times SD\ pre \times SD\ post])}$ [15], with considering coefficient correlation (r) as 0.5. Furthermore, any doubts were clarified through a discussion with the third reviewer.

Risk of bias assessment

The Cochrane Collaboration risk of bias Tool [16] was used to qualify the included RCTs based on following criteria: "randomization generation, allocation concealment, blinding of participants and outcome assessors, incomplete outcome data, selective outcome reporting and other sources of bias". Any discrepancy was discussed between the two reviewers and resolved by corresponding author.

Statistical analysis

STATA software (version 13) was used to analyze the data. Due to a high degree of heterogeneity among the included RCTs, random-effects model was used to calculate the mean difference (MD) and its 95% confidence intervals (CI). To identify statistical heterogeneity, I^2 (high $\geq$ 50%, low $<$ 50%) and P-value (significant $<$ 0.05) were used. Random--effects meta-regression was applied by mean age of participants, duration of intervention and dose of cinnamon to seek the potential sources of statistical heterogeneity between studies. Furthermore, subgroup analyses by treatment duration (2 months, 3 months), baseline SBP ($>$ 134 mm Hg, $<$ 134 mm Hg) and dosage ($>$ 1.7 gram/day, $<$ 1.7 gram/day) were performed. Also, Sensitivity analysis was undertaken to evaluate the impact of each study on the pooled results. Egger's test was applied to detect potential publication bias [17]. A $P < 0.05$ was considered as statistically significant.

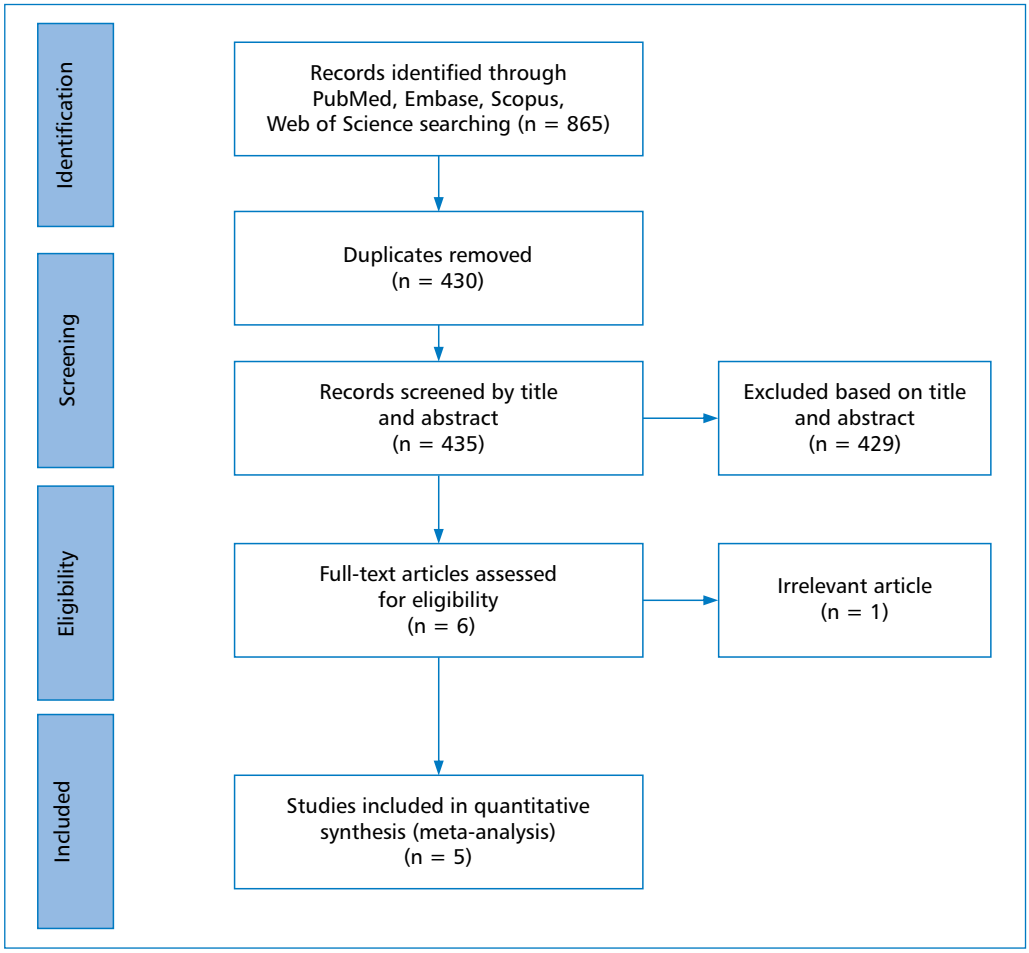

Figure 1. Flow diagram of data selection process

Results

Literature search

As shown in Figure 1, 865 papers regarding the effect of cinnamon supplementation on blood pressure were identified from online databases. Of these, 430 duplicate papers were excluded; 435 articles remained for title/abstract screening and 420 records were removed. Then, at the final step, 15 articles were assessed for eligibility and full-text screening. After full-text assessment, 5 articles were included in the present meta-analysis.

Study characteristics

Table 1 outlines the main characteristics of the included trials. Five trials were published from 2010 to 2016, which were conducted in Asia and Europe. Study sample sizes ranged from 19 to 49 participants in the intervention group and 18 to 50 subjects in the controls. A total number of participant that was included in this meta-analysis was 332. The minimum and maximum mean age of the subjects in the treatment group was 54.9–61.7 years, respectively. The duration of the interventions was 2 months in three studies and 3 months in the two other studies. The minimum dose of cinnamon supplementation among the studies was 1.2 gram/day and the maximum was 3 grams/day. All of the included trials used oral cinnamon as the supplement. The methodological quality and risk of bias of the included RCTs based on authors' judgment is shown in Table 2.

Effect of cinnamon supplementation on blood pressure

Pooled effects of 5 datasets (Figure 2) revealed a significant reduction in SBP (MD = –0.53, 95% CI = [–1.03, –0.02], P = 0.04, I^2 = 79.8%) following cinnamon administration compared with the control participants. Furthermore, high heterogeneity was decreased following subgroup analysis (Table 3) by baseline SBP (> 134 mm Hg), treatment duration (2 months) and doses less than 1.7 gram/day.

Table 1. Demographic characteristics of the included RCTs

First author (year)	Publication year	Country	Sample size (intervention/ /control)	Intervention groups age, mean (SD)	Population	Duration	Dose (gram/ /day)	Type of supplement
Akilen et al. [10]	2010	United Kingdom	30/28	54.9 (10.14)	T2DM	3 months	2	Capsule
Wainstein et al. [13]	2011	Israel	29/30	61.7 (6.3)	T2DM	3 months	1.2	Capsule
Vafa et al. [34]	2012	Iran	19/18	54.11 (10.37)	T2DM	2 months	3	Capsule
Sengsuk et al. [11]	2016	Thailand	49/50	57.2 (1.1)	T2DM	2 months	1.5	Capsule
Azimi et al. [18]	2016	Iran	40/39	54.15 (1)	T2DM	2 months	3	Powder

T2DM — type 2 diabetes mellitus

Table 2. Methodological quality assessment of the included studies

Study	Sequence generation	Allocation concealment	Blinding	Blinding of outcome assessment	Incomplete outcome data	Selective reporting	Other biases
Sengsuk (2016) [11]	+	+	+	+	+	+	+
Azimi (2016) [18]	+	+	+	−	?	+	+
Vafa et al. [34]	?	?	+	+	+	+	+
Wainstein (2011) [13]	+	+	+	+	+	−	?
Akilen (2010) [10]	+	+	+	+	+	+	+

+ — low risk; − – high risk; ? – unclear

Moreover, from the current meta-analysis we found that DBP was significantly affected by cinnamon supplementation (MD = –0.70, 95% CI = [–1.30, –0.07], P = 0.03, I^2 = 86.0%) in type 2 diabetic patients as compared with the control group. Also, high statistical heterogeneity decreased following subgroup analyses (Table 3) by baseline SBP (> 134 mm Hg), intervention duration (3 months) and doses less than 1.7 gram/day.

Meta-regression

As shown in Table 4, meta-regression of SBP and DBP could not identify the potential source of heterogeneity by mean age, duration and dose of cinnamon.

Publication bias and sensitivity analysis

No significant publication bias was found for both SBP (P = 0.60) and DBP (P = 0.78). Also, impact of each trial on the pooled result was checked and showed that the results of the present study were not affected by removing one study at a time.

Discussion

To the best of our knowledge, this is the first study to review the available literature and recent randomized clinical trials (RCTs) regarding the effects of cinnamon supplementation on blood pressure in diabetic patients as a meta-analysis. The results of this study demonstrated that cinnamon supplementation significantly reduced systolic blood pressure. But there was a high heterogeneity among studies regarding the effects of cinnamon on SBP and this was reduced by sub-group analysis and considering different features of the included studies such as duration, dose of supplementation, and baseline BP. Moreover, patients with type 2 diabetes receiving cinnamon supplement also showed significant improvements in DBP compared with the control group.

Many studies have evaluated and confirmed the hypotensive effects of cinnamon [9, 18, 19]. Our results were also in line with the results of a study by Akilen et al. as they demonstrated that cinnamon supplementation could significantly decrease SBP in diabetic patients [10]. Furthermore, our results also confirmed the results of a study by Sengsuk et al. regarding the blood pressure-lowering effects of cinnamon [11].

A possible explanation for the effects of cinnamon on BP might be related to its active components, including: cinamaldehyde, cinnomic acid, eugenol,

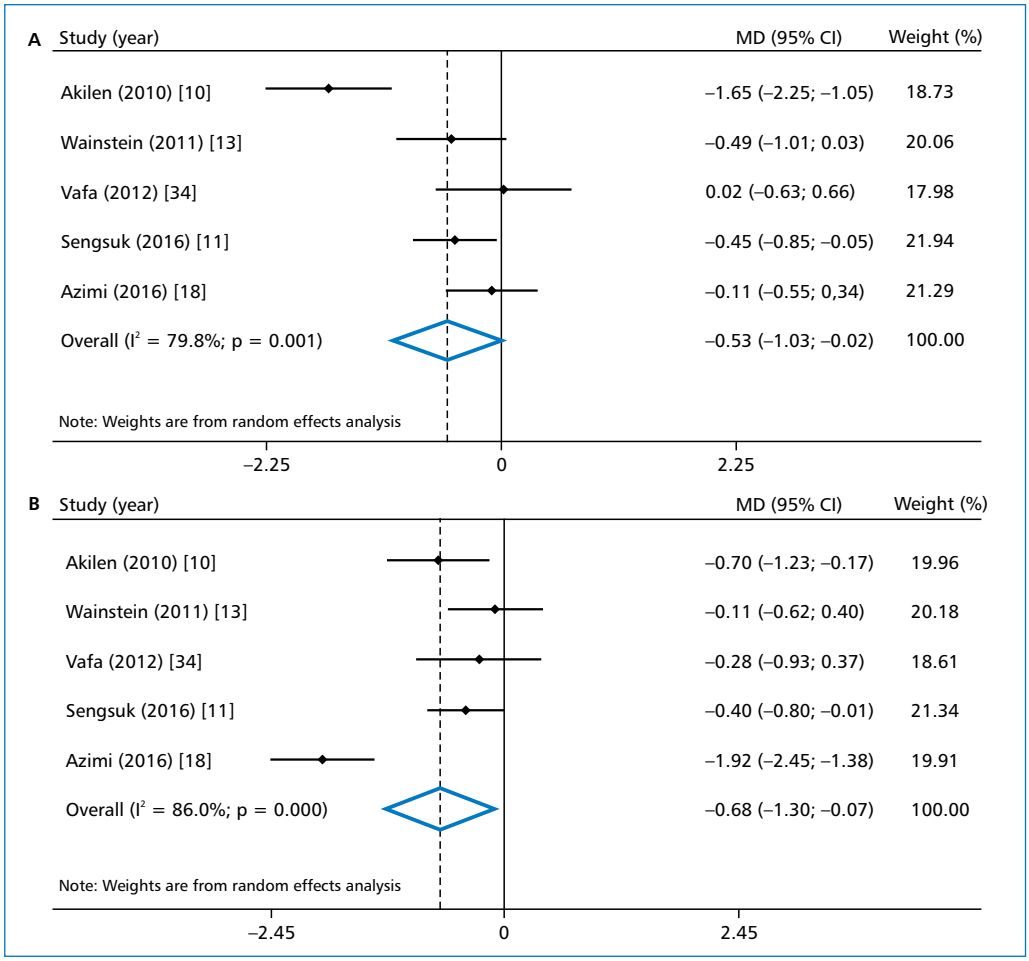

Figure 2. Forest plot detailing MDs and 95% CIs for the meta-analyses of SBP (**A**) and DBP (**B**)

and coumarin [19]. Some of the flavonoids and phytochemicals in cinnamon might cause endothelial relaxation. Cinnamon has been reported to increase the levels of cyclic guanosine monophosphate (cGMP) that can mediate the nitric oxide (NO) production and help the relaxation of vascular smooth muscle [20]. In addition, active ingredients of cinnamon might exert an anti-hypertensive effects through their cholinergic and diuretic properties [21, 22]. Another mechanism regarding the anti-hypertensive effects of cinnamon is related to its effects as an antioxidant and hypoglycemic agent that both are completely pertinent to the improvement of endothelial dysfunction [10, 23]. In diabetic patients, these effects may be more pronounced. Normal vascular contraction through normal Ca^{2+} influx via insulinotropic effects of cinnamon is the main cause of hypotensive effect of cinnamon in diabetic patients [24].

Cinnamon may also have BP-lowering properties through peripheral vasodilation [25]. Furthermore the potential beneficial effect of cinnamon may be explained by its effect on increasing estimated glomerular filtration rate. The damaging effects of hypertension may in part be due to its effects on the kidney which might be attenuated by cinnamon ingestion [26]. In one study, the effects of cinnamon on reducing vascular cell-adhesion molecule-1 and SICAM-1 was reported that was mediated through inhibiting the expression of endothelial factors at the transcriptional level especially by decreasing mRNAs [27]. Additionally, it seems that cinnamon might mediate antihypertensive effects by improving arterial wall compliance [28]. It can also enhance NO production according to one previous study [29]. Moreover, cinnamon can alleviate hyperuricemia [30, 31], resting tachycardia, nerve traffic, and high plasma norepinephrine [32], that may all be associated with better BP control.

Another important point in the current analysis is about the difference between studies regarding the cinnamon dose that was evaluated by sub-group analysis. It was shown that the hypotensive effects of cinnamon on SBP and DBP were significant at doses of < 1.7 g/day and > 1.7 g/day, respectively. Finding an exact dose of cinnamon for improving cardio-metabolic

Table 3. Subgroup analyses of SBP and DBP based on publication year, intervention duration and dose

Subgroup	Effect size	MD	95% CI	Overall P	Heterogeneity (I^2/P)
SBP					
Baseline SBP					
> 134 mm Hg	2	–0.463	–0.779, –0.147	0.004	0.0%/0.896
< 134 mm Hg	3	–0.575	–1.587, 0.437	0.265	89.9%/< 0.0001
Intervention duration					
3 months	2	–1.062	–2.201, 0.078	0.068	87.9%/0.004
2 months	3	–0.239	–0.509, 0.030	0.082	0.20%/0.367
Dose					
> 1.7 g/day	3	–0.575	–1.587, 0.437	0.265	89.9%/< 0.0001
< 1.7 g/day	2	–0.779	–0.779, –0.147	0.004	0.0%/0.896
DBP					
Baseline SBP					
> 134 mm Hg	2	–0.292	–0.606, –0.022	0.068	0.0%/0.373
< 134 mm Hg	3	–0.976	–1.940, –0.012	0.047	88.4%/< 0.0001
Intervention duration					
3 months	2	–0.400	–0.979, 0.179	0.176	59.6%/0.116
2 months	3	–0.868	–1.886, 0.151	0.095	91.3%/< 0.0001
Dose					
> 1.7 g/day	3	–0.976	–1.940, –0.012	0.047	88.4%/< 0.0001
< 1.7 g/day	2	–0.292	–0.606, 0.022	0.068	0.0%/0.373

MD — mean difference; CI — confidence intervals; SBP — systolic blood pressure; DBP — diastolic blood pressure

Table 4. Meta-regression of SBP and DBP by mean age, duration and dose

Covariates	Coefficient	P-value	95% CI	Tau^2
SBP, overall Tau^2 = 6.75				
Mean age	0.73	0.40	–6.04, 7.50	7.76
Duration	1.95	0.50	–23.46, 27.37	8.85
Dose	5.44	0.23	–21.71, 32.61	4.176
DBP, overall Tau^2 = 8.92				
Mean age	–0.24	0.66	–1.90, 1.40	11.06
Duration	–0.39	0.90	–10.42, 9.63	11.85
Dose	2.46	0.20	–2.32, 7.26	6.25

CI — confidence intervals; SBP — systolic blood pressure; DBP — diastolic blood pressure

factors may be challenging, especially because of the effects of cinnamon on inhibiting blood clotting due to its coumarin content and considering the fact that many diabetic or cardiovascular patients are receiving anticoagulants [33]. Hence, interpreting the dose-effect of cinnamon is not easy and should be interpreted with caution. Further investigations are warranted to clarify the exact safe dose of cinnamon for controlling BP.

On the other hand, effects of cinnamon on reducing SBP was more pronounced in studies done for shorter durations (in 2 months compared with 3 months). This is in accordance with the results of the study done by Sengsuk et al. [11]. It is unclear whether this is due to compliance, or tachyphylaxis.

Another point to mention is that the effect of cinnamon supplementation on SBP is more pronounced in those with higher baseline SBP, as reported by Azimi and her colleagues [18].

It is also important to mention that the results of the current meta-analysis was not determined by any single study as assessed by sensitivity analysis.

The results of the current meta-analysis pooled the available RCTs according to the effects of cinnamon on BP control. However, this study has several weakness and the results should be interpreted with caution.

It only includes five studies, and, there was high degree of heterogeneity among studies. However, the sub-group analysis considered the differences among studies including dose, duration, and baseline BP values and their effects on changes in BP following cinnamon supplementation, and another strength of the current study, was the inclusion of trials that were not restricted to a specific country or region and our results could generalize to the different ethnic groups.

Conclusion

To sum up, in this systematic review and meta-analysis, oral cinnamon supplementation could reduce BP. More RCTs with various doses are recommended to be able to draw a final conclusion. Additionally, more animal or human studies are warranted to elucidate the exact safe and effective dose of cinnamon supplement for controlling BP in cardio-metabolic or diabetic patients.

Conflicts of interest

The authors declare to have no conflict of interest.

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Insulin pump therapy and continuous systems use in adult type 1 diabetes patients — Experts' Opinion

In the 21st century, technological progress changed the therapy of patients with type 1 diabetes. Modern personal insulin pumps, and most of all continuous glucose monitoring (CGM) systems, improve the safety and comfort of life of patients with diabetes who require insulin administration. Insulin therapy with the use of CGM enables achieving a glycemic goal close to normoglycemia without an increased risk of hypoglycemia and, above all, prevents chronic complications of diabetes. This is especially important for people who are unaware of hypoglycaemia.

The reimbursement of the continuous glucose monitoring system in children and young adults < 26 years of age treated with continuous subcutaneous insulin infusion (CSII) with the use of a personal insulin pump has increased the availability of new technologies improving the safety and effectiveness of insulin therapy for patients with type 1 diabetes in Poland. **The community of diabetes health care professionals is concerned about the fact that there is no reimbursement for adults older than 26 years of age, and particularly worrying are situations when, due to the discontinuation of reimbursement, a patient with impaired hypoglycemia awareness is deprived of a therapy that provides safety in terms of health and life and improves the quality of life**. With the benefit of the patient and broadly understood social welfare in mind, we have developed the Healthcare Provision Charter "Real-Time Continuous Glucose Monitoring System (RT-CGM) in adults over 26 years of age with type 1 diabetes and hypoglycemia unawareness (i.e. the lack of prodromal symptoms of hypoglycemia) who are treated with an insulin pump". This document is the basis for starting a process aimed at reimbursing continuous glucose monitoring in patients over 26 years of age. We trust that this action, which is in line with the recent decisions of the Ministry of Health that are beneficial for diabetic patients, will become another good investment in health of Poles.

There is a consensus of opinions that the extension of CGM reimbursement will contribute to the improvement of the quality of care for patients with diabetes in Poland. A key element of qualitative changes would be guaranteed patient education services aimed at providing the patient with knowledge and skills to independently operate the CGM system, correctly calibrate

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it, interpret the direction and rate of changes in blood glucose (glycemic trends), respond to changing blood glucose values and program alarms.

Weighing the needs and opportunities, we included in the Healthcare Provision Charter adults with type 1 diabetes and impaired hypoglycemia awareness who have used insulin pump for 12 months in the last 18 months. This Expert Consensus Statement expresses special concern for adult patients with type 1 diabetes who are living with this disease for several dozen years and whose treatment with new technologies is often conditioned by a good economic status and ability to incur costs related to the use of CGM.

Clinical benefits of providing access to the CGM system and education in the use of the CGM system in adults with type 1 diabetes include the reduction of hypoglycemia and hyperglycemia episodes (which means more time spent in the glycemic target range), better quality of life and, above all, reduced risk of development and progression of acute and chronic diabetes complications. It is particularly important to reduce the frequency of severe hypoglycemia, which is a life threatening condition. Decreased rate of hypoglycemia will also contribute to the reduction of costs related to hospitalization, absenteeism and presenteeism in the workplace.

This technology not only improves the quality of diabetes care and increases the safety of insulin therapy, but also allows communication, analysis and conclusions, making e-counseling very effective. It turned out that the use of CGM and innovative telemedicine technology is possible and especially helpful for patients during the COVID-19 pandemic.

The position statement on the monitoring and treatment of type 1 diabetes in adult patients with the use of insulin pumps and a continuous glycemic monitoring system is in line with the social need and the creation of modern healthcare involving telemedicine. In the face of the global COVID-19 pandemic, the value of technologies allowing for monitoring and assessing patients from a distance is even more apparent.

Our opinion is consistent with the 2020 Guidelines on the management of diabetic patients. A position of Diabetes Poland which indicates therapeutic goals for diabetes treatment based on the use of continuous glucose monitoring systems. These guidelines also emphasize the importance of telemedicine in optimizing diabetes control.

The community of diabetes health care professionals appreciates the steps taken by the Ministry of Health to improve the quality of treatment for diabetics in Poland. Diabetes was on the list of national health priorities defined by the regulation of the Minister of Health of February 27, 2018, next to cardiovascular diseases, cancer and respiratory diseases [1]. This priority should be realized through education and increasing access of patients to diagnostic tools and methods of monitoring the progress and effects of diabetes therapy. In 2018–2020, the decisions of the Ministry of Health led to a significant improvement in the access of patients with diabetes to publicly reimbursed medical technologies and drugs.

Telephone consultations, remote blood glucose monitoring (e.g. using RT-CGM) and video consultations are the most frequently studied telemedicine interventions, each of them leading to increased access to specialist diabetes care. Unlike in cardiology, in diabetology, telemedicine solutions are rarely used in the healthcare system, despite the fact that, according to clinical trials, they translate into improved glycemic control and even reduced the risk of hypoglycaemia [2].

Increasing the availability of continuous glucose monitoring systems and relevant education in the use of CGM for patients with diabetes aged > 26 years who are treated with a personal insulin pump would constitute another positive breakthrough in diabetes care in our country.

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Inconsistency amongst the diagnostic criteria based on Ewing's tests for diagnosing cardiac autonomic neuropathy in diabetes mellitus: an under-rated issue

Cardiac autonomic neuropathy (CAN) is a common yet overlooked complication of diabetes mellitus (DM) [1]. CAN poorly correlates with specific symptoms or clinical signs implying that it frequently remains unrecognized until late in the disease trajectory [2]. Moreover, the reported prevalence of CAN varies greatly from as low as 17% to as high as 73% [3, 4]. This huge variation in the prevalence of CAN is in part due to different diagnostic criteria used to identify CAN in various trials. Cardiovascular autonomic reflex tests (CARTs) [consists of 5 heart rate (HR) and blood pressure (BP) tests] proposed by Ewing et al. [5] are considered as the gold standard tests for diagnosing CAN in DM. There are various criteria which utilizes Ewing's test for diagnosing and staging CAN. Most widely used among these criteria is the Ewing's criterion which classifies CAN into no-CAN (all tests normal or 1 test borderline), early (1 HR test abnormal or 2 borderline), definite (two HR test abnormal) and severe (2 HR test abnormal + 1 or both BP tests abnormal) CAN category [5]. This criterion is based on the theoretical conception that sympathetic dysfunction precedes parasympathetic dysfunction in diabetic CAN and hence classifies patients with borderline HR test (which examines parasympathetic dysfunction) as early or definite CAN and those with borderline or abnormal BP test (which examines sympathetic dysfunction) along with abnormal HR tests under severe category. In DM, similar to peripheral neuropathy, the autonomic neural dysfunction progresses in a length dependent fashion and the vagus nerve (longest autonomic nerve which mediates 75% of all parasympathetic activity) fibers are affected first followed by sympathetic denervation in the later stages [6]. The above discussion makes it clear that Ewing's criterion was certainly influenced by the sequential trend of autonomic dysfunction in diabetic CAN. On the contrary, some investigations have found a simultaneous occurrence of sympathetic and parasympathetic dysfunction without any chronological order for the development of CAN and on rare occasions abnormalities in BP tests may precede the abnormalities in HR tests in DM patients which makes the sequential staging of CAN by Ewing's criterion questionable [7–9]. There are various other classification criteria (based on Ewing's test) which are being used by researchers. Bellavere's criteria include only HR tests [deep breathing test (DBT), Valsalva maneuver (VM), and 30:15 ratio] into consideration and thus does not examine CAN holistically leaving the assessment of cardiac sympathetic function untouched [10]. Furthermore, CAN subcommittee of the Toronto Consensus Panel on diabetic neuropathy suggested combined examination of both CARTs and frequency domain indices of heart rate variability (low frequency power, high frequency power and LF/HF ratio) as a robust measure of CAN diagnosis. It staged CAN into early (1 positive test), definitive (2 or 3 positive test) and severe (orthostatic hypertension + one of the previous criterion) stages without contemplating the trend of

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cardiac autonomic dysfunction in DM [11]. Mendivill et al. [12] examined the presence of CAN solely by HR tests (DBT, VM, 30:15 ratio) which led to higher reported prevalence (68%) of the disease in that study. Kempler et al. [13] considered only two tests (30:15 ratio and postural drop in BP) rather than the whole battery of CART for diagnosing CAN in DM patients and reported a much lower prevalence of the disease (36%). Similarly, many such studies exists which have not considered the entire Ewing's test battery and have either assessed sympathetic or vagal dysfunction and have left undiagnosed cases (6, 15, 16). For that reason, there is huge discrepancy in the combination of CARTs and the criteria used for the diagnosis and staging of CAN which has definitely contributed to incongruity in the reported prevalence of this condition across the population. This inconsistency is an important gap in the literature which needs to be addressed because using a particular group of CARTs might lead to either many under-diagnosed or incorrectly categorized cases. A more holistic and universal criterion should be designed by future studies for early and definitive diagnosis of CAN which considers early to advance dysfunction of autonomic nervous system rather than parasympathetic to sympathetic since there is still no clear consensus on the trend of autonomic dysfunction in DM. If the future researches could develop a more accurate criteria and implement a more consistent use of the same across the studies working towards diabetic CAN, an accurate diagnosis and staging of CAN would be possible which may help clinicians in implementing timely and appropriate management strategies. Also, an universal diagnostic and staging criterion will sought out the variability in the reported prevalence of the disease and will make the comparison across different studies easier for research professionals working in the area of cardiovascular diabetology.

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