

# A Comprehensive Analysis of Glycemic Variability and Time in Ranges Assessed using CGM in Diabetic People with Microvascular Complications: A Single-Center Retrospective Real-World Evidence Study on Indian Population

**Mudit Sabharwal**

Director & Diabetologist Dharma Diabetes & Metabolic Clinic  
B-6 Greater Kailash Enclave-2, New Delhi-India

**Srivani Palukuri**

Clinical Research Scientist & Project Lead  
Real-World Evidence Department, THB  
Technology | Healthcare | Big Data Analytics 7(2) Bays  
Kinship Square, Sector 32, Gurgaon, Haryana, India

**Saurav Deka**

AVP-Real World Evidence Real-World Evidence Department, THB  
Technology | Healthcare | Big Data Analytics 7(2) Bays  
Kinship Square, Sector 32, Gurgaon, Haryana India

## ABSTRACT

**Background:** Regular assessment of glycemic variability is important to maintain good control of glycemic levels resulting delaying the onset or progression of microvascular complications. **Objective:** To investigate association of glycemic variability (GV) and time in glucose ranges measured using CGM with diabetes microvascular complications. **Methodology:** Retrospective CGM data collected for total 214 types 1 and 2 people with diabetes with (G1) and without (G2) microvascular complications. The daily glucose target range (TIR) was 70-180 mg/dL and variations in time above range (TAR), >180 mg/dL, and time below range (TBR), <70 mg/dL, considered as glucose excursions. The GV was assessed by SD, CV, MAGE, and MODD. **Results:** On average, 7 and 12% of G1 people with diabetes had spent higher time in <54 (>1%) and >250 mg/dL (>5%) glucose values. In G1, around 15% of people with diabetes had hypoglycemic events (>15 mins/day) than G2. In insulin taking people with diabetes, MAGE values were about 20 (G1) and 15% (G2) higher than their respective group averages. The logistic regression had resulted significant association of TBR-54 [OR 2.7; (95% CI 1.2-6.1)] and CV [OR 1.1 (1.0-1.1)] with complications. The risk increased 1.5 times [OR 4.1 (1.4-11.7)] with night-time and 2.7 times [OR 7.5 (2.1-26.4)] during daytime hypoglycemia ( $\geq 45$  minutes). **Conclusion:** For every 1% increase (15 minutes) of time spent in <54 mg/dL had increased 2.7 times the risk for complications. It is equally important to focus on achieving % of time spent in both target glycemic values and hyper and hypoglycemia to the extent possible.

**Keywords:** Continuous Glucose Monitoring, Microvascular Complications, Glycemic Control, Glycemic Excursion, Retrospective Study.

### KEY MESSAGE

The % of time spent in target glycemic values, as well as hyperglycemia and hypoglycemia, is equally as important. Achieving 70% of time in TIR should not be the only point of focus but maintaining % of time in TBR and TAR to the extent possible is also equally necessary

### INTRODUCTION

Good glycemic control is imperative to make sure good quality of life among those living with diabetes as compared to those without diabetes [1]. Major landmark clinical trials namely, United Kingdom Prospective Diabetes Study (UKPDS) and Diabetes Control and Complications Trial (DCCT) had shown the benefits of normal or near-normal blood glucose levels, thereby reducing the risk of microvascular complications in type 1 and type 2 diabetes mellitus [2,3]. However, studies like Action to Control Cardiovascular Risk in Diabetes (ACCORD) had shown higher mortality in people with diabetes with very tight control due to recurrent hypoglycemia [4]. These trials, when combined with evidence from other relevant studies, mandates regular monitoring of blood glucose in people with diabetes with diabetes [1].

Diabetes-related complications consist of vascular and non-vascular and are similar for both type 1 diabetes and type 2 diabetes. The vascular complications are further categorized into microvascular (neuropathy, nephropathy and retinopathy) and macrovascular (coronary heart disease (CHD), peripheral artery disease (PAD) and cerebrovascular disease). Unlike macrovascular, which share pathophysiological features with both diabetic and non-diabetic, microvascular are essentially diabetes specific. Recurrent glucose excursions, specifically chronic hyperglycemia, were reported as one of the critical factors contributing towards the onset and progression of microvascular complications [5,6]. Onset of neuropathy depends on duration of diabetes, hyperglycemia, also to an extent on genetic predisposition [7]. Neuropathy had reported to occur in 50% of long-standing type 1 and type 2 diabetes condition. In India, the prevalence had predicted as about 15-29% [8,9]. Nephropathy, characterized by raised levels of urinary albumin (proteinuria), has been reported to occur 15-40% in type 1 diabetes and 5-20% in type 2 diabetes [10]. In India, according to CURES 45 study, prevalence of microalbuminuria was 26.9% in southern region [11]. Multiple studies had reported retinopathy as anywhere from 10.3% in rural south India, to 17.6% in urban south India and 21.2% in north India [8,12,13].

The ways of capturing glycemic values have rapidly been evolving with advancement of technology. In the last few decades, there has been a paradigm shift from standard lab-based measurements to self-monitoring digital glucose meters to continuous real-time monitoring tools. Nevertheless, glycated hemoglobin (HbA1C) has been considered as a gold standard to measure long-term glycemic control for last two-three months. However, in recent times, concerns have raised on HbA1C as an inadequate indicator to capture events of glucose excursions (extent and duration), especially during night-time (nocturnal hypoglycemia). Also, the clinician (patient) must wait for three months to get the analysis done. These shortcomings surmounted in next-generation monitoring tools wherein glucose values were monitored either frequently (e.g., Self-Monitoring of Blood Glucose - SMBG) or continuously (e.g., CGM). CGM is one such sought after tool that has enticing much attention in recent times.

CGM monitors glucose values continuously and automatically in interstitial fluid for 7 to 14 days and thus generated data is retrievable at any point of time for analysis. CGM also provides a rich source of data which is further useful to calculate plethora of derivatives to draw insights analytically. Thus, captured real-time blood-glucose values would give instant and consistent visibility of patient's glucose variations to the treating clinician and if necessary, be able to intervene in the treatment.

A good number of studies had associated CGM measured glucose parameters, namely, Glycemic Variability (GV) and time in range (TIR), with diabetic complications [14-17]. Beck et al. evaluated association of percentage of time spent within and above TIR (7-point SMBG profile) with the development or progression of retinopathy and development of microalbuminuria using DCCT data [14]. They had concluded that the TIR was strongly associated with the risk of microvascular complications. Lu et al., demonstrated significant association of TIR measured using CGM with prevalence of diabetic retinopathy in about 3200 T2D people with diabetes [17]. Sartore et al., reported that GV, regardless of HbA1C, may also have a role as a risk factor for retinopathy particularly during acute fluctuations. Xu et al., study reported that GV was significantly associated with diabetic neuropathy based on a few T2D people with diabetes with tightly controlled HbA1C [17]. In a following study, conducted on large sample of people with diabetes, reported the association between increased GV assessed as MAGE (maximum amplitude of glycemic excursions) as an independent contributor to diabetic peripheral neuropathy in T2D people with diabetes [18]. Yeoh et al. in a pilot study on diabetic kidney disease (DKD) people with diabetes, reported that short-term episodic use of CGM, reduced time spent in hyperglycemia range without significantly increasing time-exposure to hypoglycemia [19]. Such comprehensive studies are scarce on Indian population, specifically diabetic people with diabetes with multiple microvascular complications. In the current study, we comprehensively analyzed association of GV and TIR assessed by CGM with diabetic microvascular complications, including neuropathy, nephropathy, and retinopathy, in type 1 diabetes and type 2 people with diabetes.

## METHODOLOGY

### Study Population

This is a retrospective, single center, observational study conducted on anonymized patient's data captured from electronic medical records (EMR) who visited the out-patient department of a clinic located in Delhi NCR, India. Total 313 diabetic patient's CGM output records in the form of ambulatory glucose profile (AGP) report were collected from December 2017 to July 2019 along with EMR clinical notes. After data cleaning and structuring, a total of 214 people with diabetes falling between 22 and 85 years of age with type 1 diabetes (n=4) and type 2 diabetes (n=210) were selected for final analysis. The remaining 99 people with diabetes were excluded from the study due to incomplete number of CGM days (<12 days) and non-availability of required clinical notes details.

Based on presence or absence of microvascular complications, the people with diabetes were categorized as -

1. Group 1 - with neuropathy, nephropathy, and retinopathy – 107 people with diabetes
  - a) Neuropathy - 67
  - b) Nephropathy - 20
  - c) Neuropathy + Nephropathy - 16

- d) Neuropathy + Retinopathy - 3
- e) Neuropathy + Nephropathy + Retinopathy - 1
- 2. Group 2 – without microvascular complications - 107

The HbA1C values were measured either using ‘standard lab methods’ or ‘spot test’ within  $\pm 0$  to 30 days around CGM initiation date. Details of medication, risk factors (dyslipidemia, hypertension, cardiac and hypothyroidism) and lifestyle (smoking and alcohol) along with clinical and demographic particulars of people with diabetes were taken from the corresponding EMRs.

### **Assessment of diabetic neuropathy, nephropathy (reduced kidney function) and retinopathy**

Diabetic peripheral neuropathy was assessed by monofilament test and vibration perception threshold (VPT) using a biothesiometer. Nephropathy was assessed from laboratory parameters like serum creatinine (0.9 to 1.3 mg/dL), moderately increased microalbuminuria (urine albumin:creatinine ratio - ACR 30-300 mg/dL). Retinopathy was assessed by an ophthalmologist with dilated comprehensive eye examination.

### **CGM Parameters**

The 24 hours CGM glucose profiles were measured using Abbott Freestyle Libre Pro device for 13 ( $\pm 1$ ) consecutive days. The filament of the CGM system was put in just under the skin of the arm on day 1 and removed after 13 ( $\pm 1$ ) days, generated daily record of 96 continuous sensor values for every 15 minutes time interval per day and 1344 records per 14 days. People with diabetes were on their daily diabetic medication, regular daily activities and diet during the CGM period. The resulted CGM data was reported in the form of AGP in line with Advanced Technologies and Treatments for Diabetes (ATTD) CGM consensus defined format [20]. The time in target range (TIR) was defined as the percentage of time spent with in the target glucose range of 70-180 mg/dL during a 24-h period. Any variation in time above range, 180 mg/dL (TAR) and time below range, 70 mg/dL (TBR) were considered as glucose excursions. Hypoglycemia was further classified as low (<70-54 mg/dL) and very low (<54 mg/L). Hypoglycemia was also analyzed in terms of number of events, mean duration of an event and % time spent in TBR. Hyperglycemia was graded as high (180-250 mg/dL) and very high (>250 mg/dL). The ATTD guidelines on % of time spent in target TIR (Table 3 from reference 20) for assessment of glycemic control for adults with type 1 diabetes and type 2 diabetes were used to analyze the AGP data [20]. The guidelines have given the ‘% of time per day’ whereas we have taken average % of time over CGM days, i.e., 13 ( $\pm 1$ ) days. Due to this reason, some patient’s % of time per day rather may vary from the average CGM values and so as TBR and TAR values. A total of 94 derivative parameters (from AGP data) were calculated using a web-based tool, GlyCulator 2.0 [21]. The resulted 94 parameters were compared between the groups (*Supplementary information*). The GV parameters included as standard deviation (SD), coefficient of variation (CV), glucose excursions as MAGE, mean of daily differences as MODD, hourly glucose changes as CONGA (1-6)h, glycemic risk assessment as GRADE.

### **Statistical Analyses**

Normally distributed data were expressed as mean ( $\pm$ SD), skewed data in median (range) and categorical data as percentages. While calculating the percentages, the respective category/group total was taken as base. The differences in continuous data or categorical data

between the two subgroups were compared using Student's T-test or the chi-square test as appropriate.

The association between CGM parameters and microvascular complication was analyzed using multivariate binary logistic regression analysis. The presence and absence of a complication was defined as 1 and 0, respectively. The continuous variables, TIR, TBR and TAR were converted into categorical variables by applying ATTD guidelines (Table 3 from reference 20). For example, when % of time was  $\geq 70\%$  in TIR was defined as '0' and  $< 70\%$  as '1' and similarly for TBR,  $< 54$  mg/dL ( $> 1\%$  - '1' and  $\leq 1\%$  - '0'), 54-70 mg/dL ( $> 3\%$  - '1' and  $\leq 3\%$  - '0'), and TAR, 180-250 mg/dL ( $> 20\%$  - '1' and  $\leq 20\%$  - '0'), and  $> 250$  mg/dL ( $> 5\%$  - '1' and  $\leq 5\%$  - '0'). The resulted odds ratio with 95% CI were reported.

All statistical analyses were performed using R program 3.6.0 and SPSS version 25 (free trial).

## RESULTS

### Characteristics of clinical variables

The clinical characteristics of study population were summarized in Table 1. As mentioned earlier, people with diabetes were categorized as group 1 (G1) and group 2 (G2) based on presence and absence of microvascular complications, respectively. Out of 214, 4 people with diabetes were type 1 diabetes and rest were type 2 diabetes, amongst, 107 people with diabetes were having diabetic microvascular complications (G1) and remaining 107 without complications (G2). After comparing the clinical parameters between the two groups, age, % of male, duration of diabetes, % of HTN, cardiac disorders and CKD were higher in G1. While, % of dyslipidemia, hypothyroidism, and smoking were comparable between the groups. The oral antidiabetic medications, metformin and sulfonyl urea were almost equally prescribed between the groups whereas DPP4i, SGLT2i and glitazones were higher in G1 than G2 (Table 1). The people with diabetes in G1 received higher insulin, including long and rapid-acting, and GLP-1 analogues than G2. Also, hypertensive and lipid-lowering drugs were higher in G1. Most of the people with diabetes (93.5%) had completed CGM for the intended duration, i.e., 14 days, and the remaining (6.5%) could only continue for 12 or 13 days.

**Table 1-Demographic and clinical characteristics of study population categorized based on with (G1) and w/o (G2) microvascular complications (N=214)**

| S. No                         | Parameter Name          | G1<br>N=107    | G2<br>N=107   | Total<br>N=214 |
|-------------------------------|-------------------------|----------------|---------------|----------------|
| <b>Demographic Parameters</b> |                         |                |               |                |
| 1                             | Type 1 diabetes (n, %)  | 2 (1.9)        | 2 (1.9)       | 4 (1.9)        |
| 2                             | Type 2 diabetes (n)     | 105 (98.1)     | 105 (98.1)    | 210 (98.1)     |
| 3                             | Male (n, %)             | 64 (59.8)      | 53 (49.5)     | 117 (54.7)     |
| 4                             | Female (n, %)           | 43 (40.2)      | 54 (50.5)     | 97 (45.3)      |
| 5                             | Age in years (mean, SD) | 57.4 (13.2)    | 46.7 (12.8)   | 52.1 (14.0)    |
| 6                             | HbA1C % - n, mean (SD)  | 106, 8.4 (1.7) | 95, 8.3 (1.8) | 171, 8.4 (1.8) |

|                                                                                                               |                                            |                |               |                 |
|---------------------------------------------------------------------------------------------------------------|--------------------------------------------|----------------|---------------|-----------------|
| 7                                                                                                             | Diabetes duration in years<br>n, mean (SD) | 70, 12.4 (9.2) | 52, 9.2 (7.8) | 122, 11.1 (8.8) |
| <b>Risk factors (n, %)</b>                                                                                    |                                            |                |               |                 |
| 8                                                                                                             | Hypertension                               | 35 (32.7)      | 19 (17.8)     | 54 (25.2)       |
| 9                                                                                                             | Cardiac (CAD and CVD)                      | 10 (9.3)       | 7 (6.5)       | 17 (7.9)        |
| 10                                                                                                            | Dyslipidemia                               | 52 (48.6)      | 56 (52.3)     | 108 (50.5)      |
| 11                                                                                                            | Hypothyroidism                             | 19 (17.8)      | 18 (16.8)     | 37 (17.3)       |
| <b>Lifestyle (n, %)</b>                                                                                       |                                            |                |               |                 |
| 12                                                                                                            | Smoking                                    | 7 (6.5)        | 9 (8.4)       | 16 (7.5)        |
| 13                                                                                                            | Alcohol                                    | 8 (7.5)        | 28 (26.2)     | 36 (16.8)       |
| <b>Medication details (n, %)</b>                                                                              |                                            |                |               |                 |
| <b>Oral antidiabetic</b>                                                                                      |                                            |                |               |                 |
| 14                                                                                                            | Metformin                                  | 105 (98.1)     | 103 (96.3)    | 208 (97.2)      |
| 15                                                                                                            | Sulphonyl urea                             | 88 (82.2)      | 83 (77.6)     | 171 (79.9)      |
| 16                                                                                                            | DPP4i                                      | 100 (93.5)     | 86 (80.4)     | 186 (86.9)      |
| 17                                                                                                            | SGLT2i                                     | 63 (58.9)      | 53 (49.5)     | 116 (54.2)      |
| 18                                                                                                            | $\alpha$ -Glucosidase inhibitor            | 4 (3.7)        | 4 (3.7)       | 8 (3.7)         |
| 19                                                                                                            | Glitazones                                 | 13 (12.1)      | 5 (4.7)       | 18 (8.4)        |
| 20                                                                                                            | GLP-1 analogues                            | 4 (3.7)        | 1 (0.9)       | 5 (2.3)         |
| <b>Insulin</b>                                                                                                |                                            |                |               |                 |
| 21                                                                                                            | Long acting                                | 45 (42.1)      | 42 (39.3)     | 87 (40.7)       |
| 22                                                                                                            | Rapid-acting                               | 18 (16.8)      | 7 (6.5)       | 25 (11.7)       |
| 23                                                                                                            | Ultra-long acting                          | 3 (2.8)        | 1 (0.9)       | 3 (1.4)         |
| <b>Concomitant Medication</b>                                                                                 |                                            |                |               |                 |
| 24                                                                                                            | Antihypertensive drugs                     | 22 (20.6)      | 10 (9.3)      | 32 (15.0)       |
| 25                                                                                                            | Lipid-lowering drugs                       | 68 (63.6)      | 56 (52.3)     | 124 (57.9)      |
| * - Patient count (n) is given for those categories where data was not available for all people with diabetes |                                            |                |               |                 |

In G1, 87 people with diabetes (81.3%) were neuropathic, 37 nephropathic (34.6 %) and 4 retinopathic (3.7%), and amongst, 16 people with diabetes (14.9%) were both neuro and nephropathic and 1 patient with all three complications (Table 2 and 3). Neuropathy was reported high percentage in female (54%) whereas nephropathy was prevalent in male (80%).

**Associations of CGM parameters with microvascular complications**

The analysis of CGM parameters was done on whole (day + night), nighttime and daytime. In general, if a mean glucose value of a patient is below or above the average, 140 mg/dL, of TIR (70-180 mg/dL), coupled with % of time (in minutes) spent, then there is a higher probability of that patient to report events of dysglycemia. Accordingly, only those parameters, which considerably varied between the groups, were only analyzed and discussed as follows, initially, on group viz., G1 and G2, then sub-group viz., (HbA1C values, risk-factors, on/off insulin), followed by logistic regression analysis results. The mean of CGM parameters is given group and category wise in Tables 2 and the normal (within ATTD guidelines) and abnormal (deviations from ATTD guidelines) % of time in TIR, TBR and TAR are depicted in Figure 1.

Group wise, the mean glucose, eA1C (GMI) and CV were comparable between G1 and G2. The GV parameters, SD, MAGE and MODD were slightly higher in G1 (Table 2). The average % of time spent in TIR was comparable between the groups, however, the number of people with diabetes with abnormal % of time in the target glucose values (20) were high in G1 when compared to G2 (Table 2 and Figure 1). About 12 and 7% of people with diabetes had spent higher time in >250 mg/dl (>1-hour 12 min/day) and <54 mg/dL (>15 min/day), respectively (Figure 1C). Approximately 5% of people with diabetes in G1 had spent <70% (<16-hours 48 min/day) in TIR (Figure 1C).

Category wise, neuropathic people with diabetes had spent on average lesser time (66%) in TIR and higher time in TBR (12.9%) and TAR (12.1%). Surprisingly, nephropathic people with diabetes, despite normal % of time (73%) in TIR, reported higher % of time (16.6% - >1-hour 12 min/day) in >250 mg/dL. While coming to multiple complications, the % of people with diabetes spent in hyperglycemia (82.3%) and hypoglycemia (76.5%) were comparatively higher than neuropathy (67.2 and 68.7%) or nephropathy (50 and 65%) alone.

Overall, about 15% more number of people with diabetes had reported hypoglycemic events in G1 than G2. Overall, the average duration of an event was longer during night (231 minutes) than daytime (180 minutes). While, in categories, multiple complication followed by neuropathy then nephropathy. However, number and duration of events were higher in neuropathy, particularly during night-time (Table 3).

The glucose excursions >1SD were measured in terms of MAGE and day wise variations as MODD were observed to be slightly higher average in G1 (by 9 and 3 units, respectively) than G2. Akin, the average MODD was nearly 3% (7 units) higher when people with diabetes suffer with multiple complications.

**Table 2-Details of demographic and CGM parameters for G1 and G2 (N=214)**

| Parameter name                       | G1<br>N=107  | G2<br>N=107  | P - Value |
|--------------------------------------|--------------|--------------|-----------|
| Male, n (%)                          | 65 (60.7)    | 52 (48.6)    | -         |
| Female, n (%)                        | 43 (40.2)    | 54 (50.5)    | -         |
| Age (in years)<br>mean (SD)          | 57.4 (13.2)  | 46.7 (12.8)  | -         |
| Diabetes duration<br>n,<br>mean (SD) | 70           | 52           | -         |
|                                      | 12.4 (9.2)   | 9.2 (7.8)    |           |
| HbA1C (%), n<br>mean, (SD)           | 106          | 95           | -         |
|                                      | 8.4 (1.7)    | 8.3 (1.8)    |           |
| Risk factors <sup>\$</sup><br>n (%)  | 76 (71.0)    | 75 (70.1)    | -         |
| <b>CGM parameters</b>                |              |              |           |
| Mean glucose (mg/dL)                 | 144.1        | 142.1        | 0.87      |
| SD                                   | 46.0         | 42.4         | 0.21      |
| Mean eA1C % (GMI)                    | 6.6          | 6.6          | 0.87      |
| <b>GV parameters</b>                 |              |              |           |
| CV - mean (SD)                       |              |              |           |
| Whole                                | 32.0 (8.0)   | 29.8 (8.2)   | 0.05      |
| Night                                | 29.5 (10.6)  | 28.0 (9.7)   | -         |
| Day                                  | 31.3 (7.8)   | 29.2 (7.9)   | -         |
| MAGE, n<br>Mean (SD)                 | 104          | 104          | 0.19      |
|                                      | 108.8 (36.8) | 99.9 (42.8)  |           |
| MODD<br>Mean (SD)                    | 38.8 (16.8)  | 35.6 (17.02) | 0.22      |
| <b>% TIR (in mg/dL)<sup>#</sup></b>  |              |              |           |
| TAR - n (%), average % of time       |              |              |           |
| >250                                 | 71 (66.4)    | 61 (57.0)    | -         |
|                                      | 11.7         | 13.2         |           |
| 180 - 250                            | 101 (94.4)   | 95 (88.8)    | -         |

|                                                                                                                                                                                                                                                                                                        |                      |                        |       |
|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------------------|------------------------|-------|
|                                                                                                                                                                                                                                                                                                        | 16.7                 | 16.3                   |       |
| Target TIR – average % of time                                                                                                                                                                                                                                                                         |                      |                        |       |
| 70-180                                                                                                                                                                                                                                                                                                 | 68.1                 | 70.7                   | -     |
| TBR - n (%), average % time                                                                                                                                                                                                                                                                            |                      |                        |       |
| 54 - <70                                                                                                                                                                                                                                                                                               | 90 (84.1)            | 86 (80.4)              | -     |
|                                                                                                                                                                                                                                                                                                        | 6.3                  | 6.1                    |       |
| <54                                                                                                                                                                                                                                                                                                    | 74 (69.2)            | 60 (56.1)              | -     |
|                                                                                                                                                                                                                                                                                                        | 4.4                  | 4.2                    |       |
| Hypoglycemic events                                                                                                                                                                                                                                                                                    |                      |                        |       |
| n (%)                                                                                                                                                                                                                                                                                                  | 62 (57.9)            | 46 (42.9)              | -     |
| average number of events                                                                                                                                                                                                                                                                               | 4.3                  | 5.6                    | 0.075 |
| Mean duration of an event (in minutes)                                                                                                                                                                                                                                                                 |                      |                        |       |
| Whole (n, %)                                                                                                                                                                                                                                                                                           | 62 (57.9)            | 46 (42.9)              | 0.31  |
| Mean (SD)*                                                                                                                                                                                                                                                                                             | 211.5                | 230.9 (182.8)          | -     |
| Median (range)                                                                                                                                                                                                                                                                                         | 176.2<br>(60-1395)   | -                      | -     |
| Night, n (%)                                                                                                                                                                                                                                                                                           | 49 (45.8)            | 39 (36.4)              | 0.42  |
| Mean (SD)*                                                                                                                                                                                                                                                                                             | 232.8                | 229.3                  |       |
| Median (range)                                                                                                                                                                                                                                                                                         | 165.0<br>(45-2430)   | 190.0<br>(45.0-1185.0) |       |
| Day - n (%)                                                                                                                                                                                                                                                                                            | 52 (48.6)            | 37 (34.6)              | 0.37  |
| Mean (SD)*                                                                                                                                                                                                                                                                                             | 182.6                | 174.6                  |       |
| Median (range)                                                                                                                                                                                                                                                                                         | 151.2<br>(60-1078.7) | 144.3<br>(60-952.5)    |       |
| \$- Hypertension, dyslipidemia, cardiac, and hypothyroidism                                                                                                                                                                                                                                            |                      |                        |       |
| * Where n values are not given consider corresponding group/category total<br>where SD was >mean value then median (range) values are given.                                                                                                                                                           |                      |                        |       |
| # <u>Note</u> : The ATTD guidelines <sup>20</sup> have given for ' % of time per day ' whereas we have<br>taken average % of time over CGM days, i.e., 12-14 days. Due to this reason, some<br>patient's % of time per day rather may vary from overall average CGM days and so<br>as for TBR and TAR. |                      |                        |       |

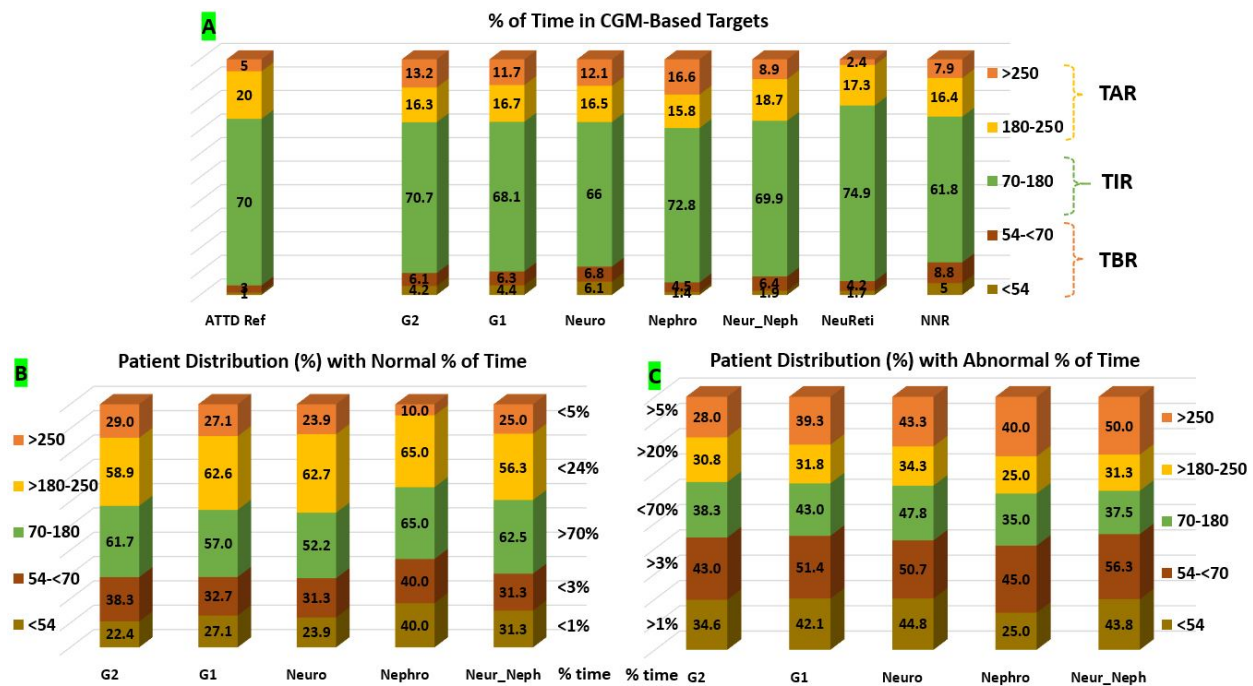


**Figure 1: A).** The % of time spent in TIR (green), TBR (brown/dark yellow) and TAR (orange/gold) across groups (G1 and G2) and categories, neuropathy (Neuro), nephropathy (Nephro), neuropathy with nephropathy (Neur\_Neph), neuropathy with retinopathy (NeuReti) and neuropathy with nephropathy and retinopathy (NNR) is given along with ATTD reference<sup>20</sup> for comparison. The % of people with diabetes with B) normal % of time (within ATTD guidelines) and C) abnormal % of time (deviation from ATTD guidelines) have given in the below panel

**Table 3-Comparison of CGM parameters across people with diabetes with single and multiple microvascular complications (N=107)**

| Parameter      | Neuropathy  | Nephropathy | Neuropathy + Nephropathy | Neuropathy + Retinopathy | Neuropathy + Nephropathy + Retinopathy |
|----------------|-------------|-------------|--------------------------|--------------------------|----------------------------------------|
| Count (n, %)   | 67 (62.6)   | 20 (18.7)   | 16 (14.9)                | 3 (2.8)                  | 1 (0.9)                                |
| Age (in years) | 58.6 (13.8) | 50.9 (14.6) | 61.3 (12.5)              | 67 (4.6)                 | 52 (0)                                 |
| Female (n, %)  | 36 (53.7)   | 4 (20)      | 2 (11.8)                 | 1 (33.3)                 | -                                      |
| Male (n, %)    | 31 (46.3)   | 16 (80)     | 15 (88.2)                | 2 (66.6)                 | 1 (100)                                |
| HbA1C, %       | 8.4         | 9.1         | 7.9                      | 7.4                      | 9.2                                    |
| eA1C, %        | 6.7         | 6.7         | 6.5                      | 6.4                      | 6.4                                    |

|                                                                                                                                                                            |                   |                 |                   |                 |       |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-------------------|-----------------|-------------------|-----------------|-------|
| Mean Glucose (mg/dL)                                                                                                                                                       | 144.8             | 145.3           | 140.9             | 135.8           | 138.3 |
| Mean SD                                                                                                                                                                    | 47.5              | 42.3            | 44.2              | 49.8            | 70.9  |
| Mean CV                                                                                                                                                                    | 32.9              | 28.9            | 31.4              | 36.7            | 51.3  |
| Mean MAGE (n)                                                                                                                                                              | 111.1             | 99.7            | (16) 106.6        | 129.8           | 132.5 |
| Mean MODD                                                                                                                                                                  | 38.4              | 36.5            | 42                | 44.9            | 76.9  |
| <b>% Time in TIR (mg/dL) - n (%), mean % of time</b>                                                                                                                       |                   |                 |                   |                 |       |
| >250                                                                                                                                                                       | 45<br>12.1 (67.2) | 10<br>16.6 (50) | 14<br>8.9 (82.3)  | 2.4             | 7.9   |
| 180-250                                                                                                                                                                    | 65<br>16.5 (97.0) | 18<br>15.8 (90) | 15<br>18.5 (88.2) | 17.3            | 16.4  |
| 70-180                                                                                                                                                                     | 66                | 72.8            | 69.4              | 74.9            | 61.8  |
| 54-<70                                                                                                                                                                     | 55<br>6.8 (82.1)  | 17<br>4.5 (85)  | 15<br>6.6 (88.2)  | 4.2             | 71.9  |
| <54                                                                                                                                                                        | 46<br>6.1 (68.7)  | 13<br>1.4 (65)  | 13<br>2.2 (76.5)  | 2<br>1.7 (66.7) | 5.0   |
| <b># Hypoglycemic events - Patient count n (%), mean events</b>                                                                                                            |                   |                 |                   |                 |       |
| Whole                                                                                                                                                                      | 39<br>5.4 (58.2)  | 9<br>2.0 (45)   | 12<br>3.0 (70.6)  | 2.0             | 8.0   |
| Night                                                                                                                                                                      | 32<br>2.2 (47.8)  | 7<br>1.8 (35)   | 8<br>2.4 (47.1)   | 2<br>1 (66.7)   | 6.0   |
| Day                                                                                                                                                                        | 33<br>5.7 (49)    | 6<br>1.3 (30)   | 11<br>1.9 (64.5)  | 2<br>1.5 (66.7) | 2.0   |
| <b>Mean duration of an event (in minutes)</b>                                                                                                                              |                   |                 |                   |                 |       |
| Whole                                                                                                                                                                      | 226.1             | 179             | 196.0             | 165             | 208.1 |
| Night                                                                                                                                                                      | 269.9             | 148.9           | 177.9             | 157.5           | 177.5 |
| Day time                                                                                                                                                                   | 199.3             | 172.5           | 152.7             | 101.2           | 112.5 |
| Note: No people with diabetes with nephropathy with retinopathy and only retinopathy; The n (%) is given when the count is less than the respective category total (N=107) |                   |                 |                   |                 |       |

### HbA1C association with CGM parameters

In a sub-group analysis of people with diabetes with respect to HbA1C values, the mean glucose values increased with increase in HbA1C values in both the groups. The time spent in TIR was

gradually decreased with increased HbA1C (except HbA1C class - 9.0-9.9) in G1 whereas no such trend was observed in G2 (Table 4).

Further analysis based on HbA1C targets, namely, stringent (<6.5%), flexible (6.5-7.5%), high (7.6-8.5%) and very high (>8.5%), evidently inferred that an overlap of people with diabetes who are meeting the targeted glycemic values (70% time in TIR) and at the same time having experienced glycemic fluctuations, especially hypoglycemia (Table 4). People with diabetes had experienced hypoglycemic episodes, on average spending >15 minutes per day, while maintaining stringent (<6.5%) or flexible (6.5-7.5%) HbA1C targets, especially in G1 than G2. When HbA1C was high (7.6-8.5 or >8.5%), had hyperglycemia besides considerable hypoglycemic events. These trends were comparable between G1 and G2.

**Table 4-Patient distribution across four HbA1C groups with abnormal % of time in hypoglycemia (TBR), hyperglycemia (TAR) and TIR for both G1 (N=107) and G2 (N=107)**

| HbA1C (%)<br>Glucose values*<br>(mg/dL) | <6.5        |             | 6.5-7.5      |              | 7.6-8.5      |              | >8.5         |              |
|-----------------------------------------|-------------|-------------|--------------|--------------|--------------|--------------|--------------|--------------|
|                                         | G1          | G2          | G1           | G2           | G1           | G2           | G1           | G2           |
| Count, n                                | 11          | 16          | 26           | 22           | 22           | 23           | 46           | 35           |
| <b>Hyperglycemia (TAR), n (%)</b>       |             |             |              |              |              |              |              |              |
| >250                                    | 2<br>(18.2) | 1<br>(6.3)  | 2<br>(7.7)   | 3<br>(13.6)  | 10<br>(45.5) | 4<br>(17.4)  | 25<br>(54.3) | 17<br>(48.6) |
| 180-250                                 | 1<br>(9.1)  | 3<br>(18.8) | 2<br>(7.7)   | 2<br>(9.1)   | 11<br>(50.0) | 4<br>(17.4)  | 20<br>(43.5) | 19<br>(54.3) |
| <b>TIR, n (%)</b>                       |             |             |              |              |              |              |              |              |
| 70-180                                  | 2<br>(18.2) | 4<br>(25.0) | 6<br>(23.1)  | 3<br>(13.6)  | 10<br>(45.5) | 6<br>(26.1)  | 28<br>(60.9) | 21<br>(60.0) |
| <b>Hypoglycemia (TBR), n (%)</b>        |             |             |              |              |              |              |              |              |
| 54-70                                   | 4<br>(36.4) | 6<br>(37.5) | 17<br>(65.4) | 10<br>(45.5) | 9<br>(40.9)  | 12<br>(52.2) | 19<br>(41.3) | 12<br>(34.3) |
| <54                                     | 4<br>(36.4) | 4<br>(25.0) | 8<br>(30.8)  | 6<br>(27.3)  | 3<br>(13.6)  | 9<br>(39.1)  | 15<br>(32.6) | 12<br>(34.3) |

A cursory look at eA1C values had given the impression that when values were below the group (10% TIR interval) average, then those people with diabetes had spent higher time in TBR but not in TAR. For example, 25 people with diabetes had reported eA1C in the range of 3.5-5.5% and their average TIR - 65.9%, TBR - 32.2% and TAR - 1.8% (*Supporting information*).

### Association of risk factors with CGM parameters

The two groups were further analyzed based on presence and absence of risk quarter (RQ), hypertension, dyslipidemia, cardiac, and hypothyroidism, along with the risk factors as

mentioned in ATTD guidelines (20) and >65 years age group (Figure 2). About 70% (n=151) of people with diabetes in both groups had either one or more risk factors. The microvascular complications were higher in hypertension (65%), cardiac (59%), followed by dyslipidemia (47%) and hypothyroidism (51%) when compared to no-RQ people with diabetes (49.2%) (Table 5). Gender wise, male population with RQ (83, 55%) was higher than female (68, 45%) out of which, higher number of male (46/83, 55%) were with microvascular complications than female (30/68, 44%). Both RQ and no-RQ subgroups had considerable amount of GV, but surprisingly, the variations were better controlled in RQ people with diabetes than no-RQ, particularly, TIR, TBR, TAR, MAGE, MODD and number of hypoglycemic events (Table 5).

**Table 5-Patient distribution (n, %) is given for subgroups RQ and w/o RQ in group (N=214)**

| Parameter                                  | RQ           |              |           |             | no-RQ     |
|--------------------------------------------|--------------|--------------|-----------|-------------|-----------|
|                                            | Hypertension | Dyslipidemia | Cardiac   | Hypothyroid |           |
| Total                                      | 54           | 108          | 17        | 37          | 63        |
| G1                                         | 35 (64.8)    | 51 (47.2)    | 10 (58.8) | 19 (51.4)   | 31 (49.2) |
| <i>Abnormal % of time in target ranges</i> |              |              |           |             |           |
| TIR, n (%)                                 | 12 (34.3)    | 19 (37.3)    | 4 (40)    | 10 (52.6)   | 14 (45.2) |
| TBR, n (%)                                 | 6 (17.1)     | 13 (25.5)    | 1 (10)    | 3 (15.8)    | 15 (48.4) |
| TAR, n (%)                                 | 6 (17.1)     | 16 (31.4)    | 2 (20)    | 8 (42.1)    | 13 (41.9) |
| Mean MAGE (mg/dL)                          | 95.9         | 105.5        | 84.9      | 104.3       | 118.2     |
| Mean MODD (mg/dL)                          | 31.6         | 36.8         | 29.6      | 37.5        | 43.8      |
| <i>Hypoglycemic events, n (%)</i>          |              |              |           |             |           |
| Whole                                      | 21 (60.0)    | 28 (54.9)    | 5 (50)    | 9 (47.4)    | 21 (67.7) |
| Nighttime                                  | 17 (48.6)    | 20 (39.2)    | 4 (40)    | 7 (36.8)    | 20 (64.5) |
| Daytime                                    | 15 (42.9)    | 23 (45.1)    | 3 (30)    | 8 (42.1)    | 20 (64.5) |

Approximately, 25% of G1 and 6% of G2 were of >65 years with averages 73 and 70 years, respectively (Figure 2). The % time in TIR for both the groups were within ATTD suggested ranges for older age, albeit with higher % of time in TBR.

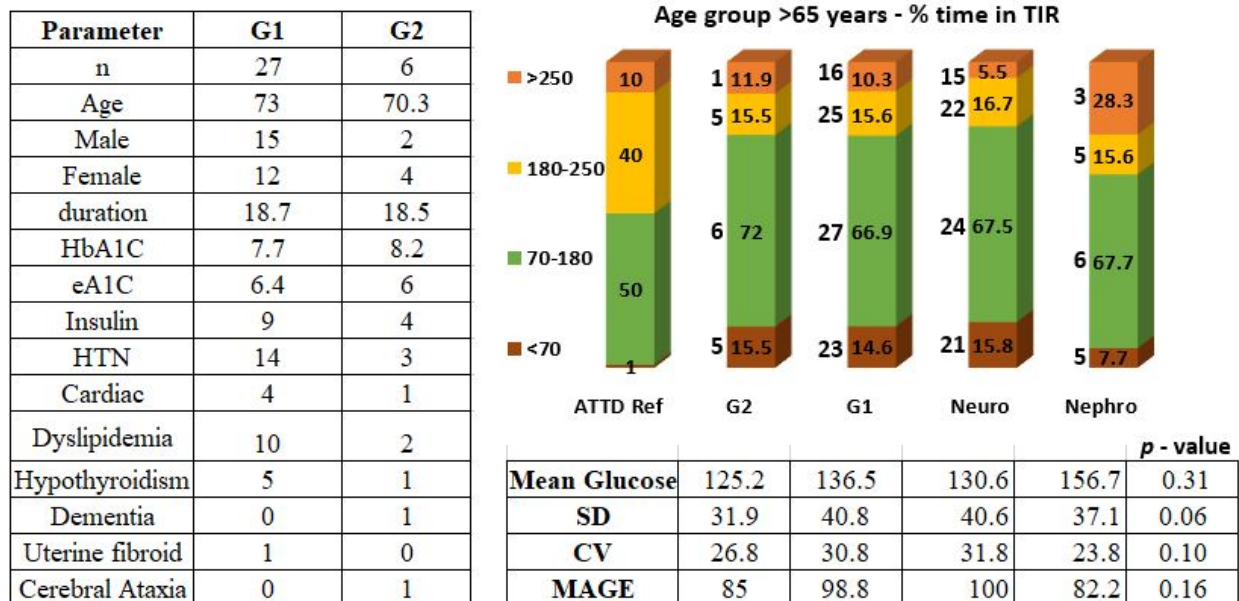


**Figure 2: Risk-patient characteristics of (>65 years) G1 and G2 are shown in columns in right-side panel. The average % of time in TIR of people with diabetes with >65 years age for G1, G2, neuropathy and nephropathy categories are given on the left above panel. The count of people with diabetes is given next to each corresponding TIR class and glucose variables are provided below for respective groups and categories along with *p*-value**

### Insulin as monotherapy or in combination (with oral antidiabetic)

A subgroup of insulin taking people with diabetes in both groups, G1 and G2, were analyzed across properties shown in Table 6. The duration of diabetes, HbA1C, eA1C, mean glucose, SD, CV, MAGE, MODD were higher in insulin taking subgroups. However, the subgroup having microvascular complications had shown relatively higher glucose variations as reflected in parameters namely, mean glucose, SD, CV, MAGE and MODD. Also, about 18 and 9% more number of people with diabetes had spent in >250 and <54 mg/dL, respectively than the corresponding G2 subgroup. The MAGE values of insulin subgroup (129.4 and 114.8) were about 18.7 and 12.9% higher than their respective group averages (109 and 100 – Table 2), respectively. The mean duration of a hypoglycemic event was higher in insulin subgroups for both G1 and G2.

**Table 6-Comparing demographic and CGM parameters across people with diabetes taking insulin**

| Parameter                                     |                            | G1           |                 | G2           |                 |
|-----------------------------------------------|----------------------------|--------------|-----------------|--------------|-----------------|
|                                               |                            | With insulin | Without insulin | With insulin | Without insulin |
| Count                                         |                            | 49           | 58              | 42           | 65              |
| Age (in years), mean (SD)                     |                            | 57.9 (12.4)  | 57.7 (13.3)     | 47.9 (14.4)  | 46.0 (11.6)     |
| Male, n (%)                                   |                            | 28 (57.1)    | 36 (62.1)       | 18 (42.8)    | 35 (53.8)       |
| Female, n (%)                                 |                            | 21 (42.8)    | 22 (37.9)       | 24 (57.1)    | 30 (46.2)       |
| Duration of diabetes (in years), (n), average |                            | (35) 16.0    | (36) 8.7        | (26) 10.2    | (26) 8.6        |
| HbA1C (%) (n), average                        |                            | 9.1          | (56) 7.8        | (37) 9.3     | (59) 7.6        |
| <b>CGM parameters (mg/dL) - (n), average</b>  |                            |              |                 |              |                 |
| Mean glucose                                  |                            | 164.9        | 126.4           | 151.6        | 135.9           |
| SD                                            |                            | 56.3         | 37.4            | 49.7         | 37.7            |
| CV                                            |                            | 34.9         | 29.6            | 33.3         | 27.5            |
| eA1C (%)                                      |                            | 7.4          | 6.0             | 6.9          | 6.4             |
| MAGE                                          |                            | 129.4        | (57) 91.8       | (40) 114.8   | (64) 90.6       |
| MODD                                          |                            | 48.5         | 30.6            | 41           | 32.2            |
| <b>% Time in TIR (mg/dL) - (n), average</b>   |                            |              |                 |              |                 |
| >250                                          |                            | (44) 15.3    | (27) 5.9        | (30) 15.0    | (31) 11.6       |
| 180-250                                       |                            | 21.2         | (52) 12.4       | (39) 30.4    | (57) 20.6       |
| 70-180                                        |                            | 58.2         | 76.4            | 61.8         | 76.4            |
| 54-70                                         |                            | (40) 4.6     | (50) 7.6        | (36) 11.7    | (51) 7.0        |
| <54                                           |                            | (35) 4.3     | (39) 4.6        | (26) 5.6     | (35) 3.2        |
| Hypoglycemia                                  | # events                   | (31) 3.7     | (31) 5.0        | (22) 7.7     | (24) 3.6        |
|                                               | Mean duration (in minutes) | 232.4        | 190.6           | 219.2        | 241.6           |

## Type 1 People with diabetes

As mentioned earlier, there were four type 1 people with diabetes, two each in G1 and G2, without any reported comorbidities and risk-factors. The average mean glucose was 216.6 and 167.1 mg/dL and % of time in TIR, 30.0 and 39.2%, respectively. The GV parameters were highly fluctuated as reflected in SD – 81.0 and 89.1, CV – 37.4 and 53.4 and MAGE – 190.9 and 227.0, for G1 and G2, respectively.

## Multivariate Logistic Regression Analysis

In a multivariate binary logistic regression model with G1 and G2, significant associations were observed with CGM parameters when included as categorical variables. The analysis results are given in Table 7. The parameters, TBR-54 (<54 mg/dL) [OR 2.7 (95% CI 1.2-6.1), *p-value* <0.05] and CV [OR 1.1 (1.0-1.1), *p-value* <0.05] came significant along with age [OR 1.1 (1.0-1.1), *p-value* <0.05], gender [OR 1.8 (0.9-3.4), *p-value* 0.08], HbA1C [OR 1.2 (0.9-1.5), *p-value* 0.2] insulin [OR 1.2 (0.6-2.6), *p-value* 0.6] and RQ [OR 1.2 (0.6-2.5), *p-value* 0.6]. Further analysis on TBR-54 as function of time of the day during prolonged hypoglycemic event (≥45 minutes) had shown that the risk was increased by 1.5 times [OR 4.1 (1.4-11.7), *p-value* 0.01] in presence of hypoglycemic events at night and 2.7 times [OR 7.5 (2.1-26.4), *p-value* <0.05] at daytime (Table 7).

**Table 7-Determinants of microvascular complications as positive outcome using binary multivariate logistic regression analysis**

| Parameter                                                                                   | B   | S.E. | Wald | OR  | 95% CI |       | p-value |
|---------------------------------------------------------------------------------------------|-----|------|------|-----|--------|-------|---------|
|                                                                                             |     |      |      |     | Lower  | Upper |         |
| Age                                                                                         | 0.1 | 0.0  | 32.2 | 1.1 | 1.1    | 1.1   | <0.05   |
| Gender                                                                                      | 0.6 | 0.3  | 3.0  | 1.8 | 0.9    | 3.5   | 0.08    |
| HbA1C                                                                                       | 0.2 | 0.1  | 2.0  | 1.2 | 0.9    | 1.4   | 0.16    |
| Insulin                                                                                     | 0.2 | 0.4  | 0.3  | 1.2 | 0.6    | 2.6   | 0.57    |
| RQ                                                                                          | 0.2 | 0.4  | 0.3  | 1.2 | 0.6    | 2.5   | 0.60    |
| CV                                                                                          | 0.1 | 0.0  | 5.7  | 1.1 | 1.0    | 1.1   | 0.02    |
| TIR-Night                                                                                   | 0.4 | 0.4  | 1.4  | 1.6 | 0.7    | 3.3   | 0.24    |
| TBR-54                                                                                      | 1.0 | 0.4  | 5.4  | 2.7 | 1.2    | 6.1   | 0.02    |
| <b>Risk enhancement of TBR-54 in presence of prolonged hypoglycemic event (≥45 minutes)</b> |     |      |      |     |        |       |         |
| Eq 1 +<br>Hypoglycemic event at night                                                       | 1.4 | 0.5  | 6.9  | 4.1 | 1.4    | 11.7  | 0.08    |
| Eq 1 +<br>Hypoglycemic event at daytime                                                     | 2.0 | 0.6  | 9.8  | 7.5 | 2.1    | 26.4  | 0.02    |

Insulin – people with diabetes on insulin vs off insulin; RQ – hypertension, cardiac, dyslipidemia and hypothyroidism; TBR-54 – patient's % of time in <54 mg/dL glucose values; TIR-night – Patient's % of time in target TIR (70-180 mg/dL) during nighttime.

Eq 1 – age + gender + HbA1C + Insulin + RQ + CV + TIR-Night + TBR-54

## DISCUSSION

In the current study, we have investigated the association of GV parameters assessed using CGM, specifically focused on glucose excursions, and other parameters with microvascular complications in diabetic people with diabetes. The CGM data generated for 13 ( $\pm 1$ ) days in terms of 94 derivative parameters were correlated with groups with and w/o microvascular complications. Our analysis had provided the following important insights - Comprehensive analysis of GV and other standard parameters in association with microvascular complications of age groups from 22 to 85 years. The percentage of people with diabetes spent abnormal % of time in TIR/TAR/TBR were comparatively high in the complication group. The % of time spent in TBR-54 was a significant contributor to complications wherein for every 1% increase (15 minutes) of time spent in <54 mg/dL, 2.7 times increased risk for complications. The duration and occurrence time of the event considerably augments (2.7 times in daytime and 1.5 times in night-time) the extent of the risk. Also, age and gender were significant contributors ( $p$ -value <0.05) whereas HbA1C, risk-factors (comorbidities) and insulin taking (higher/uncontrolled glucose values) were not significant ( $p$ -value >0.05) contributors.

The glucose excursions (hyperglycemia) have been proven to bring in pathological changes in microvessels by multiple mechanisms that led to a range of functional irregularities. Microvessels, the smallest functional component of the cardiovascular system, distinct from macrovessels in architecture and cellular components, and are made up of arterioles, capillaries, and venules. They play crucial roles in regulating blood pressure and nutrients distribution which in turn managed by metabolic need generated myogenic vasomotion and permeability. According to few studies, the irregularities were reported to arise well before onset of hyperglycemic events [22,23]. Glycemic excursion driven pathological changes produce advanced glycation end products (AGEs) and increased oxidative stress via glycated proteins. The membrane and cellular proteins of insulin-insensitive cells, which are reliant on microvasculature supply, namely, Schwann cells in peripheral nerves, mesangial and podocytes in neurons and renal glomerulus, and endothelial cells (ECs) and pericytes in retina. When exposed to these by-products, these cells would undergo nonenzymatic glycosylation followed by membrane thickening. The microvascular functions of thus impaired cells would reduce eventually manifesting into clinical problems like neuropathy, nephropathy and retinopathy. Also, under aggravated conditions, promotes secondary events like, PAD, elevated hypertension, macrovascular atherosclerosis, reduced myocardium vascularization, and poor wound healing.<sup>[24]</sup>

Attaining optimal glycemic levels is imperative not only to prevent the onset but also to delay the progression of diabetic associated microvascular complications. The benefits of timely and sustained glycemic control in reducing the risks of these complications have well been established in type 2 people with diabetes [2,3]. According to UKPDS study, 1% reduction in mean HbA1C was associated with 37% reduction in microvascular complications, 14% in

myocardial infarction and 21% in diabetes-related mortality [3]. Notably, according to Wong *et al.*, microvascular complications do not occur at an arbitrary glycemic threshold [25].

Conventionally, HbA1C is the gold standard to assess the average glucose excursions (hyperglycemia) happened in last 2-3 months. However, it may not provide complete information about the acute or minute yet significant excursions (hypoglycemia) and their magnitude and frequency, particularly, during nocturnal time which may otherwise go unnoticed. On the other hand, real-time CGM data for 14 days, which has been established to correlate strongly with three months mean glucose, time in ranges and hyperglycemia events, emerging as an alternative monitoring tool [26,27]. The CGM measured eA1C (GMI) values, though have been reported to differ from lab measured HbA1C for the reasons mentioned in Johnson *et al* paper, yet repeatedly provides important insights about both higher and lower glucose levels. In the current study, a sub-group of 57 people with diabetes whose eA1C (GMI) was <5.5%, about 95% (54) people with diabetes had commonly reported weakness with lethargy and/or were suffering with chronic or serious ailments, hematuria, fibroids, DKA, or underwent PTCA etc., and their HbA1C was in a range of 5.5 to 11.4% (average 7.5%) [28]. Therefore, lower (below average) eA1C values, especially with normal or high HbA1C, might be an indicative of weakness with lethargy due to any chronic ailment or any unreported reason and should be monitored closely. Also, advised not to target to reduce HbA1C incessantly as their daily glucose levels might be under control than the values related to measured HbA1C. On contrary, higher eA1C values (>7.5%) correlated well with HbA1C and higher % of time (>1 hour 12 minutes) in TAR (>250 mg/dL).

The significant % people with diabetes (79%) on insulin were noticed to be with reduced eA1C values when compared to the corresponding lab measured HbA1C values. This was also in consistent with reported observations by Hu *et al* (2018). The reasons might be that these people with diabetes have maintained the medication and lifestyle throughout the duration without any considerable deviations or taking insulin without missing the doses. As it was mentioned earlier, this study was based on real-world data where people with diabetes could continue their daily routine lifestyle with regular medications close to suggested timings. Compared to pre-designed studies, in real-world evidence studies, the conditions were not well controlled and hence, have the advantage of capturing the patient conditions closer to everyday activities. This indeed provide a better understanding and estimation of a treatment, medication and diagnostic tools used under ambulatory (real-world) conditions. Thus, eA1C, particularly when (far) less than the respective HbA1C in insulin taking patient, can be a useful indicator to assess the extent of insulin-linked hyperglycemia.

### **Risk factor's role in microvascular complications**

Risk for a diabetic patient can be of two types, risk of diabetes on existing ailments or vice versa. The first type is beyond the scope of the study. Focus was given on the second type as briefly discussed as variability in % of time in risk-individuals and its contribution as assessed by multivariate analysis. According to ATTD guidelines, high-risk individuals are those with cognitive deficits, renal disease, joint disease, osteoporosis, fracture, cardiovascular disease and elderly with >65 years age.<sup>[20]</sup> In the diabetic perspective, 'risk' was defined as severe hypoglycemia than hyperglycemia[20]. Accordingly, the % of time in the target ranges was adjusted to less stringent targets than the normal diabetic population.

Due to limitation of sample size, independent risk contribution of individual complications could not be done. According to multivariate analysis, presence of hypertension, dyslipidemia, cardiac and hypothyroidism would bring 20% risk [OR 1.20 (0.6-2.5), *p-value* 0.6], however it was not significantly differentiated (*p-value* 0.6) between the people with diabetes having and not having microvascular complications. Thus, one possible inference is presence of comorbidities contribution, if not to the progression, might be to the onset of microvascular complication.

Hypertension is a well-known risk contributor to diabetes kidney disease, nephropathy and retinopathy. The uncontrolled blood pressure makes kidney arteries narrow and harden, which eventually damage the kidney and its function. Similarly, damages blood vessels in retina creating pressure on optic nerve and subsequently compromises the vision. A recent study on type 1 diabetes proposed nerve conduction velocity was associated with hypertension and systolic BP [29]. In our study, 65% (35) of people with diabetes with hypertension (n=54) had reportedly nephropathy and/or neuropathy, and amongst, 22% (12) with abnormal % of time in TIR. However, it is non-significant (*p-value* 0.9) contributor with OR 1.0 (95% CI 0.4-2.4). Dyslipidemia, proven risk-factor of diabetes, was also reported to contribute to onset and progression of microvascular complications. Deranged lipid values along with visceral fat upregulate inflammatory mediators which subsequently induce microvasculature. Interestingly, in our study, the people with diabetes with dyslipidemia were in good control of glycemic parameters when compared to off people with diabetes. Moreover, dyslipidemia came as insignificant risk factor (*p-value* 0.7) in our analysis. The results (hypertension and dyslipidemia) might be confounded by the usage of antihypertensive and lipid-lowering medications due to which BP and lipids were under control.

### **Insulin role in complications**

Out of available antidiabetic therapeutic options, insulin is one of the treatment of choices for effective long-term control of glycemic levels, especially, for people with diabetes with severe  $\beta$ -cell dysfunction. The DCCT study and Kumamoto trial had undeniably established the role of insulin in slowed or delayed onset of retinopathy, nephropathy and neuropathy in type 1 diabetes and type 2 diabetes, respectively [2,30]. In our study, those people with diabetes who were put on insulin had (on average) high HbA1C values (>9.0%). The subgroup analysis has revealed that higher mean glucose values with increased HbA1C. This observation is in correlation with reported study by Vigersky and McMahon (2019) [31]. Hypoglycemia is the after-effect commonly reported in insulin taking people with diabetes and the same was observed in our study wherein the variations higher than 1SD as measured by MAGE in insulin taking subgroups. The MAGE values of subgroups were about 18.7 and 12.9% higher than their respective group averages. The logistic regression also showed that the insulin increases the risk of complications by 24% (*p-value* 0.6). In another way, uncontrolled long-term high glycemic values are known contributors to complications.

### **Hypoglycemia**

Hypoglycemia, via oxidative stress, induces myocardial infarction, cardiac arrhythmias, ischemic stroke and ischemic cerebral damage. The acute and longitudinal events of hypoglycemia may perhaps be an added risk factor to cardiovascular incidences, cognitive impairments and other comorbidities, and vice versa. The nocturnal events are of concern than the episodes during daytime for the very reason of going unnoticeable and often followed by

morning hyperglycemia. A post hoc analysis of first 7 days CGM parameters vs last 7 days (data not shown) had revealed that the group average % time in TAR (>180 mg/dl) of people with diabetes was effectively controlled (first week - 25.5% to last week - 16.7%) albeit, with increased % time in TBR (7.9% to 11.2%). The parameter TBR-54 (OR 2.7) resulted as 160% times risk factor towards complications. The risk was 1.6 times [OR 4.3 (95% CI 1.6-11.5), *p-value* <0.05, data not shown] in presence of hypoglycemic events for  $\geq 45$  minutes. This explains that the duration of the event alongside the occurrence time of the event appears to be deciding factors for how much risk being contributed to the progression of complications. Further to justify, TBR-54 risk was increased 1.5 times (Table 8) with night-time and 2.7 times (Table 8) during daytime prolonged hypoglycemic event ( $\geq 45$  minutes). For the reason, additional precaution should be taken while targeting for tight glycemic control particularly for vulnerable people with diabetes with multiple complications.

### LIMITATIONS

This being a cross-sectional study having an inherent limitation to analyze temporal cause-and-effect relationship of the above-discussed insights. Also, the study sample was collected from one center located in Delhi-NCR region hence, extendibility of results is rather limited. Importantly, we have not considered different stages of severity of microvascular complications to study the association of disease status wise changes with observed insights. The sample sizes of nephropathy and retinopathy are not enough to make conclusive observations. However, our study insights are meaningful and useful when combined as a overall microvascular complications group.

### CONCLUSION

Good number of studies have undeniably proven the importance of rigorous glycemic control to attain multiple benefits, essentially, to delay the onset and progression of microvascular complications. The factors like old age, duration of diabetes along with risk-factors like hypertension, dyslipidemia, cardiac and renal problems are add-on burdens which make the glycemic targets rather difficult to achieve and adhere. Nevertheless, it become quite arduous and challenging task for clinicians to strike a balance between hypoglycemia and hyperglycemia while attaining ideal % time (70%) in targeted TIR. It is apparently important to have a close monitoring of a vulnerable patient who is expected to show variations. In such situations, CGM is a tool of choice, which enables real-time monitoring with a check on blood glucose excursions (hypoglycemia) especially during nocturnal hours. CGM parameters such as TIR can be considered as an alternative monitoring parameter to HbA1C with an added benefit in capturing acute and minute yet significant excursions. Real-world data studies on diabetic people with diabetes with the aid of 24x7 monitoring tools like CGM are need of the hour to establish not merely the real-time evidence, also to enable doctor-patient relation in finding problematic areas and subsequently to set achievable goals. Considering the significant role of glucose excursions in onset of microvascular complications, it is important to have a consistent check of dysglycemia in no-complications group as well.

Our study has provided a comprehensive analysis of 90 plus derivative parameters generated using CGM for 13 ( $\pm 1$  day) subsequent days in 214 diabetic people with diabetes with and without microvascular complications. The complications group was observed to spent higher time, >1 (>15 minutes) and >5% (1 hour 12 minutes), in <54 and >250 mg/dL, respectively. The TBR-54 i.e., hypoglycemia resulted as a significant risk factor for microvascular

complications. Importantly, the duration and occurrence time of the event has a considerable effect (2.7 times in daytime and 1.5 times in night-time hypoglycemic events ( $\geq 45$  minutes)) on the extent of the risk. In insulin taking subgroups, MAGE values were about 18.7 and 12.9% higher than their respective group averages (109 and 100) for both G1 and G2, respectively.

Our study provided a detail understanding of patterns of glycemic variations in people with diabetes with microvascular complications. The study results have undeniably proven that achieving 70% of time in TIR should not be the only point of focus but maintaining % of time in TBR and TAR to the extent possible is also equally necessary. Furthermore, helpful in carefully designing interventions targeting people with diabetes with and without complications. In addition, provided a good basis to formulate a pressing need to consider “% time in TIR” as an alternative marker to capture events that usually go unnoticed and difficult to measure using conventional tools/markers. However, further longitudinal studies on representative samples are required to ascertain these observations.

### ACKNOWLEDGEMENTS

All authors have equally contributed towards the study hypothesis, design, conduct and manuscript writing.

The work abstract (#810164)) was published in AACE Annual Scientific & Clinical Congress – May 2020, Washington, DC., Endocrine Practice 2020; 26(Supp 2):146-147.

### References

- [1]. Thomas, N., Kapoor, N., Velavan, J., & Vasan, K. S. (2018). Chapter 9. *Gadgets and glucose monitoring: A practical guide to diabetes mellitus*. Jaypee Brothers Medical Publishers Pvt.
- [2]. The effect of intensive treatment of diabetes on the development and progression of long-term complications in insulin-dependent diabetes mellitus. *New England Journal of Medicine*, 329(14), 977–986.
- [3]. UK Prospective Diabetes Study (UKPDS) Group. (1998). Intensive blood-glucose control with sulfonylureas or insulin compared with conventional treatment and risk of complications in patients with type 2 diabetes. *Lancet*, 652, 837–853.
- [4]. Action to Control Cardiovascular Risk in Diabetes Study Group, Gerstein, H. C., Miller, M. E., Byington, R. P., Goff, D. C., Bigger, J. T., Buse, J. B., Cushman, W. C., Genuth, S., Ismail-Beigi, F., Grimm, R. H., Probstfield, J. L., Simons-Morton, D. G., & Friedewald, W. T. Effects of intensive glucose lowering in type 2 diabetes. (2008). *New England Journal of Medicine*, 358(24), 2545–2559.
- [5]. Powers, A. C., Stafford, J. M., & Rickels, M. R. (2018). Chapter 398. *Diabetes Mellitus Complications: Harrison's Principles of Internal Medicine 2nd volume Twentieth Edition*, 2. McGraw-Hill Education.
- [6]. Orasanu, G., & Plutzky, J. (2009). The pathologic continuum of diabetic vascular disease. *Journal of the American College of Cardiology*, 53(5) Suppl., S35–S42.
- [7]. Fowler, M. J. (2008). Microvascular and macrovascular complications of diabetes. *Clinical Diabetes*, 26(2), 77–82.
- [8]. Chawla, A., Chawla, R., & Jaggi, S. (2016). Microvascular and macrovascular complications in diabetes mellitus: Distinct or continuum? *Indian Journal of Endocrinology and Metabolism*, 20(4), 546–551.
- [9]. Bansal, D., Gudala, K., Muthyala, H., Esam, H. P., Nayakallu, R., & Bhansali, A. (2014). Prevalence and risk factors of development of peripheral diabetic neuropathy in type 2 diabetes mellitus in a tertiary care setting. *Journal of Diabetes Investigation*, 5(6), 714–721.
- [10]. Gross, J. L., de Azevedo, M. J., Silveiro, S. P., Canani, L. H., Caramori, M. L., & Zelmanovitz, T. (2005). Diabetic nephropathy: Diagnosis, prevention, and treatment. *Diabetes Care*, 28(1), 164–176.

- [11]. Unnikrishnan, R. I., Rema, M., Pradeepa, R., Deepa, M., Shanthirani, C. S., Deepa, R., & Mohan, V. (2007). Prevalence and risk factors of diabetic nephropathy in an urban South Indian population: The Chennai Urban Rural Epidemiology Study (CURES 45). *Diabetes Care*, 30(8), 2019–2024.
- [12]. Rema, M., Premkumar, S., Anitha, B., Deepa, R., Pradeepa, R., & Mohan, V. (2005). Prevalence of diabetic retinopathy in urban India: The Chennai Urban Rural Epidemiology Study (CURES) eye study, I. *Investigative Ophthalmology and Visual Science*, 46(7), 2328–2333.
- [13]. Raman, R., Rani, P. K., Reddi Rachepalle, S., Gnanamoorthy, P., Uthra, S., Kumaramanickavel, G., & Sharma, T. (2009). Prevalence of diabetic retinopathy in India: Sankara Nethralaya Diabetic Retinopathy Epidemiology and Molecular Genetics Study Report 2. *Ophthalmology*, 116(2), 311–318.
- [14]. Beck, R. W., Bergenstal, R. M., Riddlesworth, T. D., Kollman, C., Li, Z., Brown, A. S., & Close, K. L. (2019). Validation of time in range as an outcome measure for diabetes clinical trials. *Diabetes Care*, 42(3), 400–405.
- [15]. Lu, J., Ma, X., Zhou, J., Zhang, L., Mo, Y., Ying, L., Lu, W., Zhu, W., Bao, Y., Vigersky, R. A., & Jia, W. (2018). Association of time in range, as assessed by continuous glucose monitoring, with diabetic retinopathy in type 2 diabetes. *Diabetes Care*, 41(11), 2370–2376.
- [16]. Sartore, G., Chilelli, N. C., Burlina, S., & Lapolla, A. (2013). Association between glucose variability as assessed by continuous glucose monitoring (CGM) and diabetic retinopathy in type 1 and type 2 diabetes. *Acta Diabetologica*, 50(3), 437–442.
- [17]. Xu, F., Zhao, L. H., Su, J. B., Chen, T., Wang, X. Q., Chen, J. F., Wu, G., Jin, Y., & Wang, X. H. (2014). The relationship between glycemic variability and diabetic peripheral neuropathy in type 2 diabetes with well-controlled HbA1c. *Diabetology and Metabolic Syndrome*, 6(1), 139.
- [18]. Hu, Y. M., Zhao, L. H., Zhang, X. L., Cai, H. L., Huang, H. Y., Xu, F., Chen, T., Wang, X. Q., Guo, A. S., Li, J. A., & Su, J. B. (2018). Association of glycaemic variability evaluated by continuous glucose monitoring with diabetic peripheral neuropathy in type 2 diabetic patients. *Endocrine*, 60(2), 292–300.
- [19]. Yeoh, E., Lim, B. K., Fun, S., Tong, J., Yeoh, L. Y., Sum, C. F., Subramaniam, T., & Lim, S. C. (2018). Efficacy of self-monitoring of blood glucose versus retrospective continuous glucose monitoring in improving glycaemic control in diabetic kidney disease patients. *Nephrology (Carlton)*, 23(3), 264–268.
- [20]. Battelino, T., Danne, T., Bergenstal, R. M., Amiel, S. A., Beck, R., Biester, T., Bosi, E., Buckingham, B. A., Cefalu, W. T., Close, K. L., Cobelli, C., Dassau, E., DeVries, J. H., Donaghue, K. C., Dovc, K., Doyle, F. J., Garg, S., Grunberger, G., Heller, S., . . . Phillip, M. (2019). Clinical targets for continuous glucose monitoring data interpretation: Recommendations from the international consensus on time in range. *Diabetes Care*, 42(8), 1593–1603.
- [21]. Czerwoniuk, D., Fendler, W., Walenciak, L., & Mlynarski, W. (2011). GlyCulator: A glycemic variability calculation tool for continuous glucose monitoring data. *Journal of Diabetes Science and Technology*, 5(2), 447–451.
- [22]. Wiernsperger, N. F. (2001). In defense of microvascular constriction in diabetes. *Clinical Hemorheology and Microcirculation*, 25(2), 55–62.
- [23]. Sheetz, M. J., & King, G. L. (2002). Molecular understanding of hyperglycemia's adverse effects for diabetic complications. *JAMA*, 288(20), 2579–2588.
- [24]. He, Z., Rask-Madsen, C., & King, G. L. (2004). Pathogenesis of diabetic microvascular complications. In R. A. De Fronzo, E. Ferrannini, H. Keen & P. Zimmet (Eds.), *International textbook of diabetes mellitus*, 2 (pp. 1135–1159). John Wiley & Sons.
- [25]. Wong, T. Y., Liew, G., Tapp, R. J., Schmidt, M. I., Wang, J. J., Mitchell, P., Klein, R., Klein, B. E., Zimmet, P., & Shaw, J. (2008). Relation between fasting glucose and retinopathy for diagnosis of diabetes: Three population-based cross-sectional studies. *Lancet*, 371(9614), 736–743.
- [26]. Xing, D., Kollman, C., Beck, R. W., Tamborlane, W. V., Laffel, L., Buckingham, B. A., Wilson, D. M., Weinzimer, S., Fiallo-Scharer, R., Ruedy, K. J., & Juvenile Diabetes Research Foundation Continuous Glucose Monitoring Study Group. (2011). Optimal sampling intervals to assess long-term glycemic control using continuous glucose monitoring. *Diabetes Technology and Therapeutics*, 13(3), 351–358.

- [27]. Riddlesworth, T. D., Beck, R. W., Gal, R. L., Connor, C. G., Bergenstal, R. M., Lee, S., & Willi, S. M. (2018). Optimal sampling duration for continuous glucose monitoring to determine long-term glycemic control. *Diabetes Technology and Therapeutics*, 20(4), 314–316.
- [28]. Johnson, M. L., Martens, T. W., Criego, A. B., Carlson, A. L., Simonson, G. D., & Bergenstal, R. M. (2019). Utilizing the ambulatory glucose profile to standardize and implement continuous glucose monitoring in clinical practice. *Diabetes Technology and Therapeutics*, 21(S2), S217–S225.
- [29]. Ponirakis, G., Petropoulos, I. N., Alam, U., Ferdousi, M., Asghar, O., Marshall, A., Azmi, S., Jeziorska, M., Mahfoud, Z. R., Boulton, A. J. M., Efron, N., Nukada, H., & Malik, R. A. (2019). Hypertension contributes to neuropathy in patients with type 1 diabetes. *American Journal of Hypertension*, 32(8), 796–803.
- [30]. Ohkubo, Y., Kishikawa, H., Araki, E., Miyata, T., Isami, S., Motoyoshi, S., Kojima, Y., Furuyoshi, N., & Shichiri, M. (1995). Intensive insulin therapy prevents the progression of diabetic microvascular complications in Japanese patients with non-insulin-dependent diabetes mellitus: A randomized prospective 6-year study. *Diabetes Research and Clinical Practice*, 28(2), 103–117.
- [31]. Vigersky, R. A., & McMahon, C. (2019). The relationship of hemoglobin A1C to time-in-range in patients with diabetes. *Diabetes Technology and Therapeutics*, 21(2), 81–85.