https://doi.org/10.85475/rjd-2023-1344
Rom J Diabetes Nutr Metab Dis 2023;
www.jrdiabet.ro
Volume 30, Issue 4, pages 1344-1352

Estimated glucose disposal rate, a simple and novel index to access cardiometabolic risk among Type 2 Diabetes Mellitus patients. A Cross-Sectional Study.

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Abstract:

Objective: Insulin resistance is the common soil for Cardio vascular diseases (CVD's) and diabetes. Insulin resistance (IR) leads to atherogenic dyslipidemia characterized by increased Triglycerides, decreased HDL-C, and increased sd-LDL. Dyslipidemia leads to early predisposition of T2DM patients to CVD's. Hence, we aimed to identify and analyse the relationship between surrogate markers of IR and cardiometabolic risk biomarkers among T2DM patients.
Methods: This cross-sectional study recruited 82 T2DM without any established liver and cardiac disorders. Routine biochemical parameters were analyzed by an autoanalyzer. Index parameters like lipid pentad index, HOMA, eGDR etc. were calculated using the established formulas. HbA1C was estimated using Biorad D10 autoanalyzer. Apo-B, Apo-A, lipoprotein (a), insulin, were analyzed using ELISA.
Results: HOMA1-IR, HOMA2-IR showed a significant positive correlation with the cardiometabolic risk biomarkers. Significant negative correlation between eGDR, a surrogate of IR with the cardiometabolic risk markers in T2DM patients was observed. The median levels of CLTI, MLTI, LPI, TyG index, AIP were highest in the lowest tertile of eGDR (eGDR <4) and low in the highest tertile of eGDR (eGDR >7). We did not observe any gender specific differences in the correlation of eGDR with cardiometabolic risk biomarkers.
Conclusion: Our study was the first to demonstrate the significant negative correlation between eGDR, a surrogate of IR with the cardiometabolic risk markers in T2DM patients. These findings are important as evaluating eGDR in the routine clinical set up is easy and will help in catching the T2DM at its early phase, thus curtailing the incidences of associated comorbidities.

Keywords: Cardiometabolic risk, Estimated glucose disposal rate (eGDR), HOMA, Insulin resistance, Lipid Pentad Index, Type 2 Diabetes Mellitus.

1) INTRODUCTION:

Diabetes mellitus (DM), a disease of failed glucose homeostasis is a complex, progressive, and chronic disorder. Type 2 diabetes mellitus (T2DM) contributes to around 90% of all the DM cases in adults(1). As per the reports of World Health Organization (WHO) and United Nations, T2DM is approaching epidemic dimensions and is a major non communicable global health concern(2). According to the International Diabetes Federation (IDF) Diabetes Atlas 2019 (9th edition), the global prevalence of DM was 9.3%, this reflects a population of 463 million people with DM worldwide. The figure is projected to be 10.2% (578 million people) for the year 2030 and 10.9% (700 million people) by 2045(3). India is the epicenter of DM with 73 million patients, second largest after China as of 2017(4).

The cardinal characteristics of T2DM are insulin resistance (IR) and hyperglycemia. The chronic complications of T2DM are microvascular (neuropathy, nephropathy, and retinopathy) and macrovascular (coronary artery disease, heart failure, stroke etc.)(5). Active clinical efforts are majorly directed towards understanding the pathophysiology of T2DM to combat hyperglycemia effectively. However, Insulin resistance (IR) is the earliest manifestation of T2DM that precedes non-physiological hyperglycemia(6). Persistent hyperglycemia is the clinical symptom of T2DM, when β cells of the pancreas fail to compensate for IR. Previous studies have reported that the state of IR without diabetes is a risk factor for cardiometabolic disorders(7). Insulin being the anabolic hormone, stimulates the glucose uptake, promotes glycogen synthesis, stimulates triacylglycerol synthesis, and inhibits lipolysis. In the state of IR, defective glucose uptake and enhanced lipolysis result in glucotoxicity and

lipotoxicity, that mediate the chronic complications of T2DM(8).

Measurement of IR and considering IR as a therapeutic target for T2DM may prove helpful in curtailing the progression of TD2M. However, quantifying IR in a clinical set up is a challenge as the gold standard method, euglycemic hyperinsulinemic clamp is invasive, laborious, and impractical to be employed at the large scale(9). For these reasons, surrogate markers or indices have been developed to proxy IR measurement. The most commonly and widely employed surrogate marker for IR measurement is homeostasis model for IR (HOMA-IR)(10). The variants of HOMA modelling like HOMA1-IR, HOMA2-IR, QUICKI (marker of insulin sensitivity), have the disadvantage that their measurement involves fasting insulin estimation that limits their use in clinical setting(11,12). Moreover, HOMA modelling reflects only hepatic glucose production and not the systemic glucose uptake (majorly by the skeletal muscles) which is the earliest defect in T2DM(10,11). Insulin free index and non-steady state (dynamic) measurement of IR, led to the development of surrogate marker, estimated glucose disposal rate (eGDR). Readily available clinical parameters like waist circumference, presence of hypertension, and glycosylated haemoglobin A1c (HbA1c) make it possible to employ eGDR as a surrogate marker of IR at the population level(13). Multiple studies have established the high agreement of eGDR with the clamp technique results and validated it as a marker of T2DM complications(14,15).

Altered lipid metabolism as seen in IR is commonly referred to as diabetic dyslipidemia that is characterized by increased triglyceride (TG), decreased high density lipoprotein-cholesterol(HDL-C), and increased small dense-low density lipoprotein-cholesterol (sdLDL-C) in the circulation(16). Insulin signaling maintains the vascular tone by regulating the balance between vasoconstriction mediated via endothelin and vasodilation mediated via nitric oxide (NO) production(17). This lipid triad along with the endothelial dysfunction is a major contributor of cardiometabolic disorders in T2DM. Although there is a clear correlation between T2DM and cardiovascular diseases (CVD), their molecular pathogenesis is not well elucidated. To the best of our knowledge, only traditional lipid parameters and HOMA based IR markers are analyzed to study the relationship. Our study was planned to identify and analyse the relationship between multiple surrogate markers of IR and cumulative lipid indices reflecting cardiometabolic risk in T2DM patients. Therefore, the objective of this study was to assess IR by using surrogate markers of IR like HOMA-IR, QUICKI, eGDR and find their association with Cardiometabolic risk biomarkers [Lipid Pentad Index (LPI), Comprehensive lipid tetrad index (CLTI), Modified lipid tetrad index (MLTI), Atherogenic Index of Plasma (AIP), TyG index in T2DM patients.

2) MATERIALS AND METHODS:

2.1 Study participants:

This hospital based, single center cross sectional study recruited the patients attending the Endocrinology out-patient department (OPD) of Jawaharlal Institute of Postgraduate Medical Education and Research (JIPMER), Puducherry. The study completed all the requisite formalities and approvals of the Institute Research Council and Institute Human Ethics Committee. The study conducted all the procedures in accordance with the ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1964, as revised in 2013. Based on current criteria of the American Diabetes Association, 2012, eighty-two confirmed T2DM patients on oral hypoglycemic drugs aged between 30 to 60 years were recruited in the study. T2DM patients without any history of viral hepatitis, any form of jaundice, and established heart diseases were excluded from the study. This study did not include T2DM patients who were on lipid-lowering therapy, insulin therapy and anti-inflammatory drugs. In addition, this study did not include patients with microvascular complications [neuropathy, retinopathy, and nephropathy]. Chronic alcoholics, pregnant women and lactating women were excluded from the study.

2.2 Sample size calculation:

The sample size was estimated using the formula for testing one Correlation Coefficient. The anticipated correlation coefficient between IR and Cardiometabolic risk factors among T2DM patients was 0.35. The sample size was estimated to be 82 at 5% level of significance and 90% Power.

2.3 Clinical and Biochemical parameters:

Personal details and medical histories of all the study participants were recorded. The study participants were informed about the study and the protocol was explained to them in their vernacular. All the study participants voluntarily provided written informed consent prior to the recording of the study parameters. A single professional investigator registered the anthropometric parameters like height, weight, waist circumference, and seated blood pressure. Under strict aseptic conditions, five milliliter of blood was collected from all the study participants' antecubital veins. Serum was separated from the collected blood by centrifugation at room temperature. The collected serum was made into aliquots and one of the aliquots was sent for estimating the fasting blood glucose, lipid profile [total cholesterol (TC), triglycerides (TG's), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), very low-density lipoprotein (VLDL)] and liver function tests [Aspartate transaminase (AST), Alanine transaminase (ALT), Alkaline phosphatase (ALP), Gamma Glutamyl transferase (GGT), Bilirubin, total protein, albumin] by clinical chemistry autoanalyzer (Beckman Coulter

Au5800, Orlando, FL, USA). The rest of the aliquots were stored at -40 ºC for further analysis.
Glycated hemoglobin (HbA1c) was estimated using Biorad D10 autoanalyzer employing the principle of Ion-exchange high-performance liquid chromatography (IE-HPLC).

Serum Apolipoprotein – B, Apolipoprotein – A1, Lipoprotein (a) were analyzed by ELISA kit (Elabscience Biotechnology, USA) following manufacturer's instructions.
Serum insulin and C-peptide were analyzed by ELISA kit (Calbiotech Inc CA) following the manufacturer's instructions.
HOMA-IR, QUICKI, TyG index, AIP, CLTI, MLTI, LPI, FLDI were calculated using the established formulas(11,18–21).
HOMA2-IR, HOMA2-IR (C-peptide) were calculated using the HOMA CALCULATOR(22).
Estimated glucose disposal rate (eGDR) was calculated utilizing the composite of waist circumference (WC), hypertension (HTN) and HbA1c as:
eGDR (mg/kg/min) = 21.158 – (0.09 X WC) – (3.407 X HTN) – (0.551 X HbA1c), whereby HTN is (1 = yes, 0 = no). Systolic blood pressure (BP) of more than or equal to 140 mmHg and diastolic BO of more than or equal to 90 mmHg were considered indicators of hypertension(14).

2.4 Statistical analysis

Both descriptive and inferential statistics were used to analyze the data. Kolmogorov-Smirnov test was employed to assess the normality of continuous data. Normally distributed continuous data was represented as mean ± standard deviation (SD) and non-normally distributed data by median and inter-quartile range (IQR). Correlation between surrogate markers of insulin resistance and cardiometabolic risk biomarkers was determined by Spearman rank correlation test. Comparison of cardiometabolic risk biomarkers between the different groups was done by Kruskal-Wallis test. Cardiometabolic risk biomarkers between male and female group were compared by Mann-Whitney "U" test. Analysis was carried out at 5% level of significance and $p < 0.05$ was considered as statistically significant. Statistical analysis was performed using IBM SPSS statistics version 19.

3) RESULTS:

3.1 Baseline clinical characteristics and biochemical measurements

The descriptives of general characteristics of the study participants are shown in Table 1. Out of total 82 T2DM patients, 43 (52.4%) were males and 39 (47.5%) were females. The mean age of the study participants was 51.15 ± 6.25 years. The mean value of waist circumference of the study participants was 93.1 ± 7.42.

3.2 Correlation between surrogate markers of insulin resistance and cardiometabolic risk markers

Insulin resistance biomarkers [HOMA1-IR, HOMA2-IR, and HOMA2-IR (C-peptide) showed a significant positive correlation with Cardiometabolic risk biomarkers (LPI, CLTI, MLTI, AIP, TyG Index). Insulin resistance biomarker eGDR showed a significant inverse correlation with LPI, CLTI, MLTI, AIP, TyG Index. Similarly, Quantitative insulin sensitivity check index (QUICKI), an established surrogate biomarker of insulin sensitivity showed a significant negative correlation with cardiometabolic risk biomarkers as shown in Table 2.

3.3 Comparison of cardiometabolic risk markers in type 2 diabetes mellitus patients categorized into 3 tertiles of estimated glucose disposal rate (eGDR)

Insulin resistance among the study participants was categorized based on eGDR values. The participants were divided into Tertile 1 (eGDR < 4) (lower eGDR indicates higher insulin resistance), Tertile 2 (eGDR 4-7), Tertile 3 (eGDR >7) (higher eGDR indicates lower insulin resistance). Since the data was following the non-Gaussian distribution, the median with IQR in each of the 3 tertiles is given in Table 3. The median levels of all the cardiometabolic risk biomarkers were higher in the Tertile 1 (eGDR < 4), followed by Tertile 2 (eGDR 4-7) and lowest in the Tertile 3 (eGDR >7) (p value <0.0001).

3.4 Correlation and Comparison of cardiometabolic risk markers and eGDR between male and female group

The study participants were categorized based on gender into male and female group. We analyzed the correlation between eGDR and cardiometabolic risk biomarkers in male and female group. We did not observe any gender specific differences in the correlation of eGDR with CLTI, MLTI, LPI, TyG index, AIP (Table 4). In comparison to male group, female group had higher values of eGDR, indicative of low insulin resistance in female group (p value 0.002). The female group had lower levels of LPI, CLTI, MLTI, AIP, TyG Index however cardiometabolic risk biomarkers did not considerably differ among the two groups except for the LPI (p value 0.045) as shown in Figure 1.

3.5 Correlation between surrogate markers of insulin resistance and hepatic functioning biomarkers

HOMA2-IR showed a positive correlation with gamma glutamyl transferase (GGT) (r value 0.499) and Fatty liver disease index (FLDI) (r value0.500) and negative correlation with De-Ritis ratio and total bilirubin (r value 0.143, -0.550 respectively). eGDR showed a significant positive correlation with De-Ritis ratio and total bilirubin (r value 0.356, 0.785 respectively) and significant negative correlation with GGT and FLDI (r value -0.708, -0.716 respectively) as shown in Table 5.

4) DISCUSSION:

Insulin resistance is at the grassroot of metabolic disorders like obesity, Diabetes milletus, Non-Alcoholic fatty liver disease, Poly cystic ovarian syndrome, Chronic kidney diseases, Cardio vascular

defects(7,23,24). Insulin resistance is the state of decreased responsiveness of the insulin sensitive tissues like skeletal muscle, liver, adipose tissue towards insulin(6). In compensation to the higher demands of insulin, pancreatic β cells deteriorate over time resulting in lower levels of insulin. An analysis by Whitehall II study has shown that the state of insulin resistance precedes the clinical diabetes that is characterized by increased peripheral glucose levels(25). In the state of insulin resistance, the glucose disposal rate from the circulation is decreased and free fatty acids (FFA) release in the circulation via the lipolysis of the stored triglycerides is enhanced. Thus, early insulin resistance marked by hyperinsulinemia, hyperglycemia induced β cell failure is the key pathogenic driver for the blossoming of diabetes milletus and atherogenic dyslipidemia. Multiple studies have established insulin resistance as the independent risk factor in the development of diabetes and cardiometabolic disorders(7,8).

In our study the mean waist circumference (WC) of the study participants was 93.1 ± 7.426 (Table 1). Since the waist circumference has ethnicity and sex specificity basis, we divided the study participants into male and female groups. The mean WC in male group was 95.72 ± 5.67 and in female group mean WC was 90.21 ± 8.09. As per WHO criteria for adult Asians, both the male and female group had higher WC than the established cut off's (>90 and >80 for male and female respectively)(26). Waist circumference showed significant positive correlation with cardiometabolic risk biomarkers [LPI, MLTI, CLTI, TyG index] and insulin resistance marker HOMA1-IR. Moreover, significant negative correlation was observed with QUICKI (insulin sensitivity biomarker) and eGDR (insulin resistance biomarker) (data not shown).

As IR sets in long before the symptoms of the disease manifest, there is a need for early and accurate methods for IR measurement. The direct and gold standard method of IR measurement is euglycemic hyperinsulinemic clamp technique(9), however, this procedure is laborious, expensive, time-consuming, and also necessitates an experienced technician for constant insulin/glucose infusion and repeated blood sampling that render this procedure impractical for use in routine clinical set-up. The same limitations are with insulin suppression test and Frequently Sampled Intravenous Glucose Tolerance Test (FSIVGTT)(11). Since the above-mentioned methods are impractical and difficult to adopt at the population level, there is a need to employ surrogate markers to gauge IR in the clinical settings. In our study we employed the established surrogate markers of IR to measure IR and sensitivity in T2DM patients. IR is not only the diminished utilization of glucose by insulin sensitive tissues but also the underlying cause of main non communicable diseases like T2DM, CVD, cancer, etc. IR triggered dyslipidemia also called as diabetic dyslipidemia is characterized by lipid triad: (1) increased TG, (2) decreased HDL-C, and (3) increased sdLDL(16). These features are the root cause for atherogenic phenotype resulting in various cardiometabolic diseases (stroke, angina, coronary artery disease). T2DM patients have two- to eightfold higher CVD incidence compared to normal people, thus it is necessary to harness the potential of surrogate markers of IR to curb the incidences of cardiovascular diseases(27). The evidences derived from observational studies like Framingham study, PROCAM, WOSCOPS suggest that lipid indices are more powerful CVD risk predictors than independently used traditional lipid markers(28). The 2018 new cholesterol guidelines from the American College of Cardiology and American Heart Association emphasized on the lowering of LDL-C in CVD patients as much as possible(29), however due to the inaccuracy in the measurement of LDL-C by Friedewald equation and impracticality of employing ultracentrifugation followed by β quantification in clinical setting(30), it is recommended to measure apolipoproteins to predict CVD outcomes. Multiple population-based studies and recent guidelines have suggested to bring Apolipoproteins and non-HDL-C to the forefront as biomarkers for CVD events(29,31). We employed CLTI, MLTI, LPI, TyG index, AIP to access the cardiometabolic risk in T2DM patients. Furthermore, these consolidated lipid indices, based on emerging risk factors magnify the underlying changes in the pro/anti atherogenic lipid particles and present them as a single value for the easy evaluation. Incorporation of genetic contributor like Lp(a) in the calculation of the lipid indices adds significance to the study as Indian population has higher preponderance for CVD(32). In our study, we assessed the correlation between surrogate markers of IR and biomarkers of cardiometabolic risk in T2DM patients. Our data demonstrated the significant positive correlation of cardiometabolic risk biomarkers (CLTI, MLTI, LPI, TyG index, AIP) with IR markers (HOMA1-IR, HOMA2-IR, HOMA2-C-peptide). We also observed a significant inverse correlation of CLTI, MLTI, LPI, TyG index, AIP with eGDR (lower values of eGDR correlate with greater IR) (Table 2). These findings shed light on positive association between IR and dyslipidemia that increases the risk of CVD in diabetes patients. These observations are in concordance with most of the published data(33). Out of all the IR markers analyzed, eGDR showed a strong correlation than HOMA1-IR, HOMA2-IR, HOMA2-C-peptide with cardiometabolic risk biomarkers. Since previous studies have reported weaker correlation of HOMA-IR with clamp technique and HOMA modelling reflects only hepatic IR and not the systemic IR(34). IR seen at the level of skeletal muscle and vascular smooth muscle (as measured by clamp technique) is the causal factor for the enhanced atherosclerotic CVD not the hepatic IR, this may be reason for moderate correlation of HOMA-IR with cardiometabolic risk markers. eGDR is a validated, easy to use indirect biomarker to assess IR in T1DM and numerous studies have established its inverse correlation with micro/macro-vascular complications in T1DM(35). Data from the recent studies have revealed the potential of eGDR in assessing the IR in T2DM patients. Work done by Zelin et al, and Nystrom et al have demonstrated the association of eGDR with coagulation indices, bone turnover markers, CVD, and all-cause mortality in T2DM patients(14,15,36). Multiple reports have

suggested that the WC performs better as an anthropometric tool for the prediction of cardiometabolic risk than BMI and central obesity is better predictor of CVD than general obesity(37), therefore, we employed WC based equation for the calculation of eGDR in our study. To supplement our findings, QUICKI (marker of insulin sensitivity) showed a significant inverse correlation with cardiometabolic risk biomarkers (Table 2).

Using the cut- off values as mentioned in Table 3, our study participants were divided almost equally into three tertiles (n= 31, n=29, n=22), since there is no established cut-off for Indian T2DM patients. Based on this cut off, it was observed that the median levels of CLTI, MLTI, LPI, TyG index, AIP were highest in the lowest tertile of eGDR (eGDR <4). The median levels of CLTI, MLTI, LPI, TyG index, AIP were low in the highest tertile of eGDR (eGDR >7) and intermediate in the tertile of eGDR 4 – 7. Cumulatively, these results demonstrate that greater the IR (low eGDR value) higher is the risk for development of CVD and inverse is true with the insulin sensitivity (high eGDR value).

Since, WC has gender specific cut offs and previous studies have also shown gender specific association of eGDR with various complications in T2DM patients(15). Thus, we divided our study participants into male and female group. The median levels of eGDR were lower in male group (3.996 (3.035-4.762)) compared to female group (5.282 (4.057-9.091)). Higher levels of eGDR in female group suggest the lesser IR in female group compared to the male group (Figure 1A). This gender difference may be because of the higher estrogen levels in the female group, which were not analyzed in this study. Estrogen suppresses hepatic gluconeogenesis and is a known antioxidant thus decreasing IR in females (as reflected by high eGDR)(38). Next, we analyzed the correlation between eGDR and cardiometabolic risk biomarkers in male and female group (Table 4). In our study, we did not observe any gender specific differences in the correlation of eGDR with CLTI, MLTI, LPI, TyG index, AIP. Also, the median levels of CLTI, MLTI, LPI, TyG index, AIP were low in female group compared to male group (Figure 1B-F). Multiple clinical trials and cross-sectional studies have demonstrated the protective role of female hormones against CVD development. Estrogen therapy has been shown to decrease the proatherogenic Lp(a), increases the clearance of Apo-B and LDL-C from the circulation, and also increase the Apo-A (an important protein component of HDL-C that provides functionality to HDL particle)(39). Numerous studies suggesting the protective role of estrogen against CVD is in line with our data, however, mechanistic details are not fully elucidated.

IR is the hallmark and the main ground for the breeding of disorders like T2DM, CVD, NAFLD, PCOS etc.(40). We analyzed the correlation of surrogate markers of insulin resistance with the markers of hepatic function (De-Ritis ratio [AST/ALT], gamma glutamyl transferase (GGT), total bilirubin, Fatty Liver disease index (FLDI). eGDR better correlated with the hepatic biomarkers compared to the HOMA2-IR. eGDR showed inverse correlation with GGT and FLDI, and positively correlated with total bilirubin (Table 5). GGT is a marker of cellular oxidative stress and FLDI is a consolidated index of liver function. Inverse correlation of eGDR (low value signifies greater IR) with GGT suggests that the state of oxidative stress prevails in the presence of IR(41). Positive correlation of bilirubin with eGDR suggests the increase in bilirubin levels with the increase in insulin sensitivity (high eGDR value), as bilirubin is an endogenous antioxidant(42).

The strength of our study is that in our knowledge we are the first to report the association of eGDR with cardiometabolic risk biomarkers in T2DM patients. The lower the levels of eGDR the higher is the cardiometabolic risk. Early screening of the high-risk individuals using eGDR can be beneficial to reduce CVD events. Our study gains importance as the eGDR can be easily established as a biomarker in the clinical set up as the parameters involved in its calculation are routinely done at the grassroot hospitals. This is not the case with other surrogate markers as insulin estimation is not a routine parameter. Since eGDR measures IR and IR precedes T2DM, thus employing eGDR as screening tool can curtail incidences of diabetes and its associated complications.

There were certain limitations in our study. Firstly, the inherent factor of familial tendency for metabolic diseases can be a confounding factor. Secondly, normal participants as controls were not recruited to study the diagnostic performance of eGDR and establish its cut off value. Thirdly, validating the correlation of eGDR with cardiometabolic risk by incorporating the clinical parameters like carotid intima media thickness (CIMT) would have supplemented our observation.

5) CONCLUSION:

Our data demonstrates the significant positive correlation of surrogate markers of IR (HOMA1-IR, HOMA2-IR) with the cardiometabolic risk biomarkers (CLTI, MLTI, LPI, TyG index, AIP). Conclusively our data demonstrated the significant negative correlation between eGDR, a surrogate of IR with the cardiometabolic risk markers in T2DM patients. Moreover, eGDR can be employed as a clinical indicator to access the IR and predict cardiometabolic risk in T2DM patients irrespective of their gender. eGDR also correlated with the hepatic markers, thus it is interesting to study the eGDR in other IR associated disorders as well. These findings are important as evaluating eGDR in the routine clinical set up is easy and will help in catching the T2DM at its early phase, thus curtailing the incidences of associated comorbidities.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Approvals of the Institute Research Council and Institute Human Ethics Committee were obtained (JIP/IEC/2019/388 dated 11/12/2019). Informed

consents were obtained from all participants included in the study.

HUMAN AND ANIMAL RIGHTS

No animals were used in this study. Procedures followed were by ethical standards of the responsible committee on human experimentation (institutional and national) and with the Helsinki Declaration of 1964, as revised in 2013.

FUNDING:

This research was supported by an Intramural research grant from JIPMER, Puducherry, India (No: JIP/Res/Intramural/Phs2/2019-20 dated February 3, 2020).

CONTRIBUTION STATEMENT

Mohammad Athar: Conceptualization, Study design, Data acquisition and Curation, writing - original draft writing – review and editing. **Ramasamy Ramesh:** Conceptualization, Study design, Data acquisition and Curation, writing - original draft writing – review and editing, critical revision of draft**. Sadishkumar Kamalanathan:** Participant recruitment, data interpretation, critical revision of draft. **M. Lenin:** Data acquisition and Curation, data interpretation.

CONFLICT OF INTEREST

None of the authors have any conflicts of interest related to this article.

Table legends

Table 1: Clinical and metabolic characteristics of the study participants.

Table 2: Correlation between surrogate markers of insulin resistance and cardiometabolic risk biomarkers of the study participants.

Table 3: Comparison of cardiometabolic risk markers in type 2 diabetes mellitus patients categorized into 3 tertiles of estimated glucose disposal rate (eGDR).

Table 4: Correlation between eGDR and cardiometabolic risk biomarkers in male and female group.

Table 5: Correlation between surrogate markers of insulin resistance and hepatobiliary biomarkers of the study participants.

Figure legends

Figure 1: Comparison of cardiometabolic risk biomarkers between male and female group.

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Table 1: Clinical and metabolic characteristics of the study participants.

Parameter	N	MEAN ± SD / MEDIAN (IQR)
Glucose (mg/dL)	82	167.5 (143.5 - 211.3)
HbA1C (%)	82	9.22 ± 1.75
Waist (cm)	82	93.1 ± 7.426
Insulin (µU/mL)	82	18.43 (13.37 – 39.1)
HOMA1-IR	82	7.175 (5.12 – 19.09)
HOMA2-IR	76	2.535 (1.905 – 5.07)
C - PEPTIDE (ng/ml)	82	3.214 (2.025 – 4.785)
HOMA2%IR (c-peptide)	82	2.89 (1.85 – 4.498)
eGDR (mg/kg/min)	82	4.357 (3.357 – 7.857)
Total Cholesterol (mg/dL)	82	183.5 (163.5 – 212)
HDL-C (mg/dL)	82	42 (38 – 49)
LDL-C (mg/dL)	82	131.87 ± 38.2
TAG (mg/dL)	82	188 (134 – 275.3)
Non-HDL (mg/dL)	82	144.56 ± 38.5
AIP	82	0.647 ± 0.255
TyG index (mg/dl)	82	9.73 ± 0.62
LIPOPROTEIN-(a) (mg/dL)	82	14.91 (10.79 – 22.22)
Apo-A (mg/dL)	80	150.79 ± 15.34
Apo-B (mg/dL)	82	119.71 ± 49.76
COMPREHENSIVE LIPID TETRAD INDEX	82	10904 (5545 – 25959)
MODIFIED LIPID TETRAD INDEX	82	7913 (3583 – 21879)
LIPID PENTAD INDEX	80	427980 (174456 – 785665)

Table 2: Correlation between surrogate markers of insulin resistance and cardiometabolic risk biomarkers of the study participants.

	AIP	TyG index	CLTI	MLTI	LPI
HOMA1-IR	0.647*	0.699*	0.618*	0.629*	0.584*
HOMA2-IR	0.54*	0.56*	0.469*	0.482*	0.425*
HOMA2-IR (C-peptide)	0.304*	0.382*	0.348*	0.351*	0.388*
QUICKI	-0.647*	-0.699*	-0.618*	-0.629*	-0.584*
eGDR	-0.737*	-0.785*	-0.769*	-0.774*	-0.710*

*denotes significance at a P value of <0.05 (Spearman rank correlation) AIP: Atherogenic Index of Plasma, TyG: Triglyceride-Glucose, CLTI: Comprehensive Lipid Tetrad Index, MLTI: Modified Lipid Tetrad Index, LPI: Lipid Pentad Index, HOMA: Homeostasis model assessment, QUICKI: Quantitative insulin sensitivity check index, eGDR: estimated glucose disposal rate

Table 3: Comparison of cardiometabolic risk markers in type 2 diabetes mellitus patients categorised into 3 tertiles of estimated glucose disposal rate (eGDR).

Parameters	eGDR < 4 (n= 31) Median (Q1 - Q3)	eGDR 4-7 (n= 29) Median (Q1 - Q3)	eGDR > 7 (n= 22) Median (Q1 - Q3)	P value
CLTI	28884	9821	3431	<0.0001*

	(16818-112655)	(6428-17013)	(1988-7168)	
MLTI	24114 (14055-97634)	7381 (4822-12786)	2097 (1319-4691)	<0.0001*
LPI	842064 (510854-3250000)	341591 (185760-604799)	170527 (68041-245826)	<0.0001*
TyG Index	10.37 (9.93-10.68)	9.57 (9.29-9.78)	9.085 (8.83-9.38)	<0.0001*
AIP	0.91 (0.693-1.027)	0.615 (0.494-0.705)	0.355 (0.272-0.546)	<0.0001*

* Denotes significance at a P value of <0.05 (Kruskal-wallis test) AIP: Atherogenic Index of Plasma, TyG: Triglyceride-Glucose, CLTI: Comprehensive Lipid Tetrad Index, MLTI: Modified Lipid Tetrad Index, LPI: Lipid Pentad Index, eGDR: estimated glucose disposal rate

Table 4: Correlation between eGDR and cardiometabolic risk biomarkers in male and female group.

	Male (n = 43)		**Female (n = 39)**	
	eGDR		**eGDR**	
	r	**P**	**r**	**P**
CLTI	-0.757	<0.001	-0.788	<0.001
MLTI	-0.757	<0.001	-0.790	<0.001
LPI	-0.769	<0.001	-0.626	<0.001
TyG index	-0.770	<0.001	-0.822	<0.001
AIP	-0.698	<0.001	-0.813	<0.001

*Denotes significance at a P value of <0.05 (Spearman rank correlation) AIP: Atherogenic Index of Plasma, TyG: Triglyceride-Glucose, CLTI: Comprehensive Lipid Tetrad Index, MLTI: Modified Lipid Tetrad Index, LPI: Lipid Pentad Index, eGDR: estimated glucose disposal rate

Table 5: Correlation between surrogate markers of insulin resistance and hepatobiliary biomarkers of the study participants.

	De-Ritis ratio	**GGT**	**Total bilirubin**	**FLDI**
eGDR	0.356*	-0.708*	0.785*	-0.716*
HOMA2-IR	-0.143	0.499*	-0.550*	0.500*

* Denotes significance at a P value of <0.05 (Spearman rank correlation), GGT: gamma glutamyl transferase, FLDI: fatty liver disease index eGDR: estimated glucose disposal rate

Figure 1: Comparison of cardiometabolic risk biomarkers between male and female group

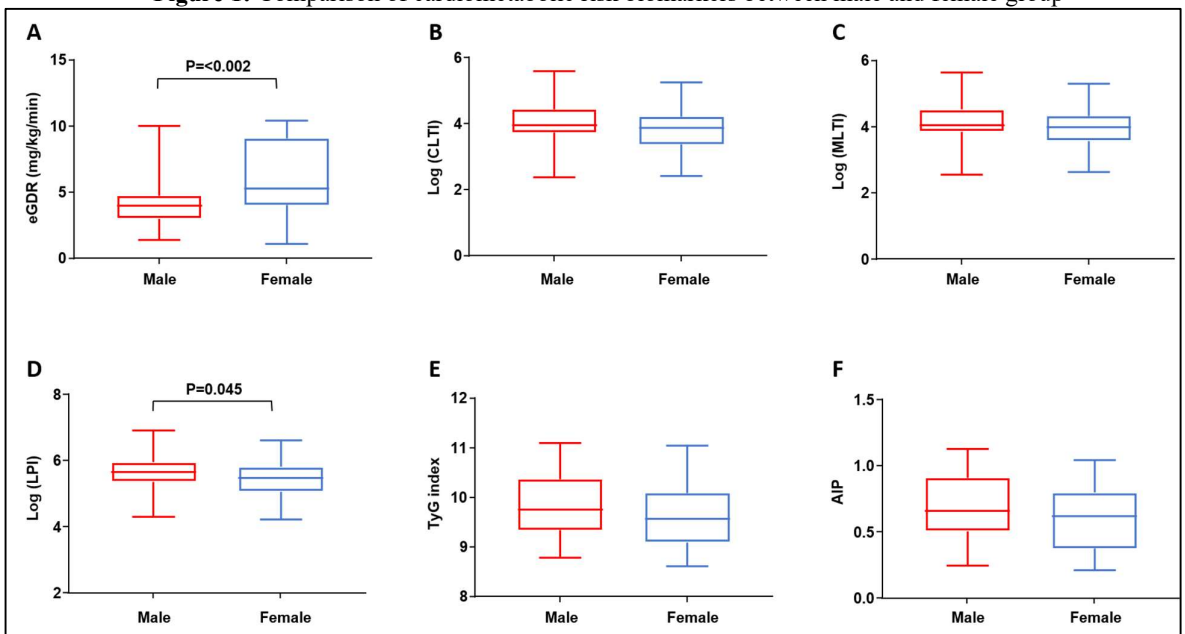

Figure 1: Comparison of cardiometabolic risk biomarkers between male and female group. Comparison of A) eGDR, B) CLTI, C) MLTI, D) LPI, E) TyG index, F) AIP between male and female group. P value of <0.05 (Mann-Whitney "U" test) was considered significant, n=43 male , n=39 female.