Role of GLYCOSYLATED Hemoglobin (HbA1c) to find out GLYCEMIC control in Type 2 Diabetes Mellitus (Controlled & Uncontrolled) in Rural Area of Western Maharashtra

[1] Dr.Geeta Ibrahim,[2] Mr. Dinkar Joshi,[3] Dr. V.S.Shindagi [4] Dr.Ashokan K.V
[1]Principle, College of Home Science Nirmala Niketan Charchget Mumbai
[2] Institute of Science Department of Biochemistry
[3] Pathologist Shindagi's Lab Sangli
[4] Prop. Department of Zoology P.V.P.College K.Mahankal Sangli

Abstract- **Glycated hemoglobin, it develops when hemoglobin, a protein within red blood cells that carries oxygen throughout the body joins with glucose in blood, becoming glycated.**

I. INTRODUCTION

Glycated hemoglobin, it develops when hemoglobin, a protein within red blood cells that carries oxygen throughout the body joins with glucose in blood, becoming glycated.

By measuring glycated hemoglobin (HbA1c) we can able to get an overall picture of our average blood sugar levels over a period of week /months.

HbA1c is also referred to as hemoglobin A1c or simply A1c. HbA1c refers to glycated hemoglobin (A1C) which identifies average plasma glucose concentration.

It can indicate people with Prediabetic, Diabetic or Normal(Normal : Below 6.0%)(Prediabetic : 6.0 to 6.4%)(Diabetic: 6.5 or above) Research has also shown that people with type 2 diabetes Mellitus if they reduces their Hba1c level by 1%.

- ❖ 19. % less likely to suffer cataracts
- ❖ 16. % less likely to suffer Heart failure
- ❖ 43. % less likely to suffer Amputation or death due to peripheral vascular disease.

In India currently diagnosed more than 62 million diabetic individuals. In 2000, 31.1 million topped the world with highest number of people with Diabetes Mellitus. According Wild et al. the prevalence of Diabetes is predicated to double globally from 171.million in 2000. To 366.million in 2030.with maximum increase in India. It is predicted that by 2030 Diabetes Mellitus may afflict up to 79.4 million individuals in India.[1,2]

Importance of Hba1c for controlling diabetes is mentioned in 1985(First time WHO report) Hba1c now use for diagnosis of diabetes it is now recommended by an international committee and ADA also. It is also useful for avoid age to age variability and also important to persons avoiding fasting and post prendial blood glucose testing. In recent year clinications strongly recommended HbA1c test for early diagnosis of diabetes mellitus.[3,4] To monitoring chronic glycemia of diabetes mellitus patients Hba1c is a gold standard method .It is clear that there is close relationship between mean blood glucose (MBG) and development of chronic diabetic complication . Prior to 1975 routine patient monitoring only to analyze glucose and ketone bodies in Urine[5] Since 1975 dramatic changes have taken place in both the methods and goals of monitoring of blood glucose. These changes were clear that hyperglycemia is responsible for diabetic complications. During the same period determination of self monitoring of blood glucose and Hba1c testing is also start (since early 1990's) [6,7]

II. BIOCHEMISTRY OF GLYCOHEMOGLOBINS:-

The non enzymatic attachment of free aldhyde groups of carbohydrates (such as glucose) to unprotonated free amino groups of proteins (such as hemoglobin) is called as Glycation.

Glycation alters the structure and function of several soluble and in soluble proteins, as well as structure and function of isolated basement membrane components. These changes are slow and cumulative resulting in long time lag between the diagnosis and progression of

complications of diabetic mellitus.[8,11]There are various molecular species in human blood, resultingfrom many potential glycation sites at the hemoglobin molecule, the different molecular former of human hemoglobin such as HbA0. (A2-b2) HbA2 (a2-d2) HbF (a2-g2) and numerous hemoglobin variants (e.g. Hbs, Hbc, HbE) Potential glycation sites of the Hemoglobin molecule includes the N-terminal amino acid Valine of the four polypeptide chains and all free C-amino groups of Lysine residues within the chains. The predominant glycation site is the N-terminal Valine residue of the b-chain of hemoglobin molecule, wich accounts for approximately 60% of all bound glucose .The term for this major component is HbA1c .other glucose molecules can bound to one or more of the 44 glycation sites of the C-amino groups within the hemoglobin molecule (34.% of all bound glucose) or at the N-terminal valine of a-chain about 6.%.[8,12,13]

There are also some further minor hemoglobin species in human blood that are adducts of other substances to hemoglobin molecule and can interfere in the determination of the GHb depending on specificity of the analytical methods. These are carbamylated and acetylated hemoglobin.

Aims and Objects:-
- ❖ To correlate HbA1C with Type II Diabetes Mellitus in controlled and uncontrolled patients.
- ❖ To study the relationship between HbA1C and controlled uncontrolled Type II Diabetes Mellitus patients
- ❖ To study the relationship between HbA1C with multiple regressive factors of Type II Diabetes Mellitus patients.(Age, Obesity, Blood Pressure, etc

III.   MATERIAL AND METHODS:-

Around 100 patients clinically diagnosed of Type II Diabetes Mellitus in age group of 30 to 60 year both male and female is recruited in OPD/IPD from following hospital.
1) Virupakash polyclinic hospital, Kavathe-Mahankal, Dist. Sangli
2) Kulkarni Hospital, Sangli.(Neurology centre)
3) Kranti Clinic Hospital, Sangli.(Cardiology Centre)

Study will be done under supervision of Dr. Vivek Shindigi M.D. Pathology and all investigations of Kavathe mahankal and Sangli hospitals are done in Shindig'slab, Sangli. Patients included in the study are categorizes into controlled/uncontrolled Type II Diabetes Mellitus by fasting /postpraridial and HbA1C.

3 ml venous blood sample is collected under all aseptic precautions. The collected blood is distributed in fluoride bulb for blood sugar and E.D.T.A. bulb for HbA1C.Sample is stored at 4-8º till further assay.

Blood sugar is done by GOD/POD colorimetric method, HbA1c is done by nyco card method.

Inclusion Criteria:-
- ❖ Patients with age group 30 to 60 year.
- ❖ Patients with one or more risk factors for developing Type II Diabetes Mellitus
- ❖ History of gestational diabetes mellitus.
- ❖ Women with PCOS
- ❖ Hypertension less than 140 to 90 mm.Hg.
- ❖ History of lipidameia and dyslipidaemia.
- ❖ History of CVD.
- ❖ Other clinical condition associated with insulin resistance e.g. severe obesity
- ❖ Patients having blood glucose above 140 and absent urine glucose.

Exclusion Criteria:-
- ❖ Pregnant women.
- ❖ Patients on chemotherapy or any anticancer drug
- ❖ HIV/HCV positive patient or patient with ART

IV.   RESEARCH DESIGN:-

This Study is done to correlate the value of HbA1C with controlled and uncontrolled Type II Diabetes Mellitus patients with fasting and post praridial blood sugar level.

Diagnostic tests of fasting plasma glucose < 130 mg/dl and post praridial plasma glucose is <180 ms/dl (with hypoglycemic drugs) is considered to be controlled type 2 diabetes mellitus patient and persons whose fasting plasma glucose> 130mg/dland postprariendial plasma glucose >250 (with hypoglycemic drugs) is considered to be uncontrolled Type II Diabetes Mellitus.

This is case controlstudy controlled Type II Diabetes Mellitus patients are control of our study and uncontrolled Type II Diabetes Mellitus patients are case of this study.FBS and PPBS were used as reference standard test.

HbA1C is measured by naycocard methods FBS and PPBS are measured by GOD/POD colorimetric method. Patients are categorized into controlled and uncontrolled

Type II Diabetes Mellitus and all parameter are comparing between this two groups Patient if positive any one of the above test is diagnosed as Diabetic.

V.   OBSERVATION AND DISCUSSION:-

We are selected age group varied from 30 to 60 year. The mean age of study group is 45 ± 15 year. Maximum number of patient is 60 in number. And age group 50-60 year. Maximum number of patient who diagnosed as uncontrolled Type II Diabetes Mellitus is also between 50-60 age groups (55%) and other 30 – 45age. (45%)

IN case of high risk patients i.e. uncontrolled Type II Diabetes Mellitus patients both male and female patients are in equal numbers. Most positive factor in our study is hypertension, high cholesterol, positive past history of obesity and genetical history of Type II Diabetes Mellitus. Out of each symptom maximum no. of patients diagnosed was polyuriea, polydysphagia, polyphagia and weakness, frequent urination, swelling on legs and some are pale and weight loss, headache, loss of appetite, some chest pane Out of100 patients, 80% patients having BMI more than normal limits and under over weight or obesity and some having central obesity and fatty. Distribution of patients study group according to FBS, PPBSL, and HbA1C with diagnostic cutoff value.

FBS mg/dl	No. of Patient	PPBS mg/dl	HbA1C%	No. of Patient
<130	41	<180=41	6.5	41
>130	59	>250=59	6.5	59
Total	100	100		100

Distribution of study group according to reference standard tests-FBS & PPBS
Controlled T2DM :(FBS < 130 & PP BS <250) total Number of patients = 41
Uncontrolled T2DM: (FBS > 130 &PPBS >250) total number of patient =59 Evaluation of HbA1C for diagnosis of Type II Diabetes Mellitus at diagnostic Cutoff value> 6.5. %
RESULT:-

HbA1C value %	Controlled T2DM	UncontrolledT2DM	Total
< 6.5	40	05	45
> 6.5	01	54	55
Total	41	59	100

HbA1c Result with respect to age group:-

Age group	HbA1c value %	Controlled T2DM	Uncontrolled T2DM
30 to 40 Year	< 6.5	10	01
40 to 50 Year	< 6.5	02	02
50 to 60 Year	< 6.5	28	02

Age group	HbA1c value %	Controlled T2DM	Uncontrolled T2DM
30 to 40 Year	> 6.5	--	04
40 to 50 Year	> 6.5	--	10
50 to 60 Year	> 6.5	01	40

Total 100 patients are studied in this research; they are distributing in to two groups.
1)   Controlled T2DM: Total patient :41 having FBS < 130 mg/dl and PPBS <
250. mg/dl.&out of 41, 40 patient having Hba1c <6.5% and 1 patients having HbA1c >6.5 %.

2)   Uncontrolled T2DM: Total patient: 59 having FBS >130 mg/dl and PPBS>250mg/dl.& out of 59, 05patient having Hba1c<6.5% & 54 patient having Hba1c > 6.5 %.

CONCLUSION:-

For evaluate Hba1c test to rule out the glycemic control in type 2 diabetes mellitus patient following measures are used at cutoff value 6.5 %.

Sensitivity = Controlled T2DM <6.5 / total ControlledT2DM × 100.   40/41× 100 = 97.56 %

Specificity = Uncontrolled T2DM >6.5 / total Uncontrolled t2DM × 100
            54/59× 100 = 91.52 %.
1)   Hba1c cut off value 6.5% showed high sensitivity & specificity to rule out the glycemic control of controlled Uncontrolled T2DM.
2)   HbA1c is Useful as the sole diabetes diabetic individuals
3)It indicates that Hba1c is in Controlled stage in older age in controlled T2Dm patients and it is uncontrolled stage, in older age, in uncontrolled T2DM patients

REFERENCES

1) Joshi SR, Parikh RM, India-diabetes capital of the world: J Assoc Physicians India. 2007,55:323-4 (Pub-Med)

2) Kumar A, Goel MK, Jain RB,Khanna P. Chaudhary,V.India Diabetes Control Key Issues, Australas med J. 2013,6(10);524-31 (PMC-free article) Pub-med

3) The value of HbA1C for diagnosis type-2 diabetes mellitus between Chinese uyghurs and Hans in Xinjians.Si-Yuan Li Si Yao wans and Guoleicao.Exp.Med.2015;8(7) 11352-11355.

4) Whiting Dr. Guariguata L,Weil C, shaw J. IDF Diabetes Atlas, Global estimate of the prevalence of diabetes for 2011 and 2030 Diabetes Res clin pract 2011,94,; 311-321 (pub-med)

5) Juarez DT, Demaris Km, Gao R, Mantza Ganian cl, Wans Smith H. Significance of HbA1C and its measurement in the diagnosis of diabetes mellitus: Us experience. Diabetes metab syndromes' 2014, 7; 487-494 (pub-med)

6) American Diabetes Association. Diagnosis, classification of diabetes mellitus. Diabetes care 2013:36 (Supp19): 567-74 (pub-med) Significance of HbA1C and its measurement in the diagnosis of diabetes mellitus; Us experience Juarez DJ et.al Diabetes metab syndrobese 2014

7) HaemoglobinA1c 1951 present and future Aes. Aldasociqi- SA, et al, Ann-Saudi med 2008 Nov-Dec.

8) Brownlee M. Negative consequences of glycation. Metabolism. 2000; 49(2) Supple 1:9–13. [Pub Med]

9) McCance DR, Dyer DG, Dunn JA, Bailie KE, Thorpe SR, Baynes JW, Lyons TJ. Maillard reaction products and their relation to complications in insulin-dependent diabetes mellitus. J Clin Invest. 1993;91(6):2470–2478.[PMC free article] [Pub Med]

10) Thorpe SR, Baynes JW. Role of the Maillard reaction in diabetes mellitus and diseases of aging. Drugs Aging.1996; 9(2):69–77. [Pub Med]

11) Vlassara H, Brownlee M, Cerami A. Nonenzymatic glycosylation: role in the pathogenesis of diabetic complications. Clin Chem. 1986; 32(10 Suppl):B37–41. [Pub Med]

12) Brownlee M, Cerami A, Vlassara H. Advanced products of nonenzymatic glycosylation and the pathogenesis of diabetic vascular disease. Diabetes Metab Rev. 1988;4(5):437–451. [Pub Med]

13) Vlassara H, Fuh H, Makita Z, Krungkrai S, Cerami A, Bucala R. Exogenous advanced glycosylation end products induce complex vascular dysfunction in normal animals: a model for diabetic and aging complications.Proc Natl Acad Sci U S A. 1992;89(24):12043–12047. [PMC free article] [Pub Med]

14) McDonald MJ, Shapiro R, Bleichman M, Solway J, Bunn HF. Glycosylated minor components of human adult hemoglobin. Purification, identification, and partial structural analysis. J Biol Chem. 1978; 253(7):2327–2332.[Pub Med]