IJBPAS, December, 2022, 11(12): 6140-6147

ISSN: 2277–4998

International Journal of Biology, Pharmacy and Allied Sciences (IJBPAS)

'A Bridge Between Laboratory and Reader'

www.ijbpas.com

DIAGNOSTIC METHODS FOR DIABETES MELLITUS

PATEL R AND SHRIVASTAV A*

Assistant Professor, Parul Institute of Applied Science, Parul University, Limda, Wagodia road, Vadodara-390025, Gujarat

***Corresponding Author: Dr. Anupama Shrivastav: E Mail: anupama.shrivastav82045@paruluniversity.ac.in**

Received 26th Jan. 2022; Revised 25th Feb. 2022; Accepted 12th March 2022; Available online 1st Dec. 2022

https://doi.org/10.31032/IJBPAS/2022/11.12.6699

ABSTRACT

Objective:

Diabetes is frequently undiagnosed. About one-third of diabetics are unaware of their condition, and the average time between start and diagnosis is seven years. The criteria for diagnosing diabetes are reconsidered in this paper, and screening criteria are recommended to make case detection easier for doctors and patients.

Subjects:

The participants in this cross-sectional study were 25 people aged 45 and up who were chosen at random.

Methods:

A total of 25 males/females who was aged between 45 to 75 were selected for the study. Midstream blood and urine were collected following the usual laboratory protocol and immediately processed in the laboratory.

INTRODUCTION

DEFINITION

Diabetes mellitus is a collection of metabolic illnesses marked by persistent hyperglycaemia caused by insulin production, insulin action, or both. The relevance of insulin as an anabolic hormone causes metabolic irregularities in carbohydrates, lipids, and proteins. These metabolic abnormalities are caused by low levels of insulin to achieve adequate response and/or insulin resistance of target tissues, primarily skeletal muscles, adipose tissue, and to a lesser extent, liver, at the level of insulin receptors, signal transduction system, and/or effector enzymes or genes. The type and duration of

diabetes determine the severity of symptoms. Some diabetic patients, particularly those with type 2 diabetes in the early stages of the disease, are asymptomatic; however, individuals with severe hyperglycaemia, particularly in children with total insulin shortage, may have polyuria, polydipsia, polyphagia, weight loss, and impaired vision **[1-4]**.

HISTORY

The phrase diabetes refers to diabetes mellitus, which is the complete name. Diabetes mellitus is a combination of the Greek word diabetes, which means "to pass through," and mellitus, which means "honeyed or sweet." This is because excess sugar is seen in both the blood and the urine of diabetics. It was dubbed the "pissing evil" in the 17th century.

Around 250 BC, Apollonius of Memphis is said to have created the word diabetes. Diabetes was first reported in English in 1425, in the form of diabetes, in the medical literature. Thomas Willis introduced the term "'Mellitus'" to the word diabetes in 1675. The pleasant taste of the urine was the reason behind this. According to their writings, the ancient Greeks, Chinese, Egyptians, Indians, and Persians recognized a pleasant taste in urine.

Diabetes was diagnosed and classed as "Madhumeha" by Sushruta (6th century BC), an Indian healer. The phrase "Madhu" means "honey" in this context, and when combined, it implies "sweet urine." An old Indian test for diabetes was to see if ants were attracted to someone's urine **[5]**.

SCREENING METHODS

Fasting blood sugar, haemoglobin (A1C), glucose tolerance tests, and random plasma sugar can all be used to screen for type 2 diabetes. Although research is being conducted to rethink this assertion, urine glucose is an inferior test for screening for diabetes.

If two abnormal tests from the same sample or two distinct samples for the first three tests below, the screening is considered positive

1) HbA1C of more than 6.5 percent

2) A fasting plasma glucose level of 125 mg/dL or higher

3) In an oral glucose tolerance test with a 75 mg glucose load, 2-hour glucose greater than or equal to 200 mg/dL.

4) In symptomatic individuals, random blood glucose levels more than or equivalent to 200 mg/dL (thirst, polyuria, weight loss, blurry vision) **[6]**.

5)Urinalysis **[5]**

Blood Glucose Levels in Fasting Plasma

An FPG test, which examines the quantity of sugar in the blood without taking into account meal consumption, is a reliable indication of glucose metabolism. It is vital to fast for at least 8 hours. The FPG and OGTT have a poor concordance; the OGTT results in more diabetes diagnoses than

either the FPG or A1c tests. In populations with particular criteria, FPG testing may be favoured above other approaches (e.g., A1c) for screening for and diagnosing diabetes (e.g., HIV). For condition-specific testing recommendations, consult the American Diabetes Association's Standards of Medical Care in Diabetes **[7]**.

The A1C test

The quantity of blood sugar (glucose) bound to haemoglobin is measured by a haemoglobin A1c (HbA1c) test. The portion of your red blood cells that transports oxygen from your lungs to the rest of your body is called haemoglobin. The average quantity of glucose connected to haemoglobin during the previous three months is determined by a HbA1c test. It's a three-month average since that's the usual lifespan of a red blood cell.

If your HbA1c readings are high, you may have diabetes, a chronic disease that can lead to major health issues like heart disease, kidney disease, and nerve damage.

HbA1c, A1c, glycohemoglobin, glycated haemoglobin, glycosylated haemoglobin is some of the other names for HbA1c **[11]**.

The A1C test determines your average blood sugar level over the previous two or three months. An A1C of less than 5.7 percent is considered normal, between 5.7 and 6.4 percent suggests prediabetes, and 6.5 percent or more implies diabetes **[8]**.

RPG (random plasma glucose) test

When diabetes symptoms are evident and health care practitioners do not want to wait until you have fasted, they may perform the RPG test to diagnose diabetes. The RPG test does not need you to fast overnight. This blood test is available at any time **[9]**.

Oral glucose tolerance test (OGTT)

Fasting plasma glucose alone fails to detect around 30% of previously undiagnosed diabetes; an OGTT is the only way to identify patients with IGT; and an OGTT is sometimes required to confirm or eliminate an impairment of glucose tolerance in asymptomatic people.

To evaluate glucose tolerance status, people with fasting plasma glucose levels of 6.1–6.9mmol/l (110–125mg/dl) should have an OGTT **[10]**.

Urine testing for diabetic analysis

Urine testing is generally inexpensive and simple to do. Urine testing can be performed to look for blood in the urine, infection (by detecting the presence of white blood cells or protein), and other systemic issues such as liver disease (by showing abnormal bilirubin levels). Ketones in the urine can also be detected by urine testing. Ketones are metabolic waste products that occur when blood glucose levels are extremely high. Ketones in the urine suggest that the patient's blood glucose level is likely to be very high, and that they may be suffering from ketoacidosis, a potentially life-threatening

complication of diabetes that requires immediate care **[12]**.

MATERIALS AND METHODS

HbA1c TEST

MATERIAL:

- Gloves
- HbA1c test cartridge
- EDTA blood collecting tube
- 1ml blood

METHOD:

- Before you began testing, put on some gloves.
- Remove the hba1c test cartridge from the refrigerator and set it aside for 10 minutes to got to room temperature before opened it and removed it by handed, being careful not to touched the lowered section.
- It should be placed on a table.
- Use the full sample straight from the tube and vigorously mixed it 8-10 times before used it.
- After removing the tube cap, removed the sampled device from the cartridge.
- Insert the sampled device into the sampled material, then observe as the blood filled the capillary while avoiding air bubbles and extra sample outside the capillary.
- As quickly as feasible, replaced the sampled device in the capillary, and at that time tapped the patient's icon to move the sample. Manually shut the covered after carefully placed the cartridge inside.
- Enter the patient's number after selected the patient's id.
- The test results were available in three minutes.

Figure 1: Sample took from tube in sampling device capillary

Blood sample collection:

venepuncture should be used to collected around 2ml of the patient's blood into a tube containing a 1:2 (w/w) combination of ethylenediaminetetraacetic acid and sodium fluoride. Two millilitres of blood require only five milligrams of the combination. To ensured complete mixing, shook the tube vigorously.

Anticoagulant mixture preparation:

A blender should be used to combine 100 mg of EDTA and 20 mg of sodium fluoride into a fine powder. This should be done under a fume hood if possible. It's best to keep the mixture in a clean container.

REAGENTS:

- 2N Sodium hydroxide (NaOH) - 8g of NaOH is dissolved, then the volume is brought up to 100ml with purified water.

- Sodium Sulphate-Zinc Sulphate Reagent: Dilute 55mL zinc sulphate solution (10g/100mL ZnSO4.7H2O) in 1 litre sodium sulphate solution (93mmol/litters).
- pH-7.2 phosphate buffer (0.05M)
- Glucose oxidase reagent: Make this reagent from scratch by combining 25 mg of glucose oxidase with 1% ortho-toluidine in sodium phosphate buffer. To 250ml of buffer, add a tiny amount of peroxidase (2mg) and makeup. If stored in a brown-coloured container at 4°C, this solution will last for around 4 weeks.

METHOD:

Pipette 0. 1 millilitres of blood into 1. 8 millilitres of sodium sulphate-zinc sulphate reagent in a centrifuge tube. Added 0. 1ml 2n sodium hydroxide, centrifuge for 5 minutes at 3000rpm, and collected 0. 5ml supernatant in duplicate.

prepare the blank with 0. 5 ml of distilled watered.

standard preparation: prepare a 200mg/dl glucose standard, then used 0. 5ml of a variety of glucose solutions (50mg/dl, 100mg/dl, 150mg/dl, and 200mg/dl) that had been adequately diluted from the standard.

• 50 mg/dl - 125 litres glucose standard + 375 litres distilled watered

• 250 l glucose standard + 250 l distilled watered - 100 mg/dl

• 150 mg/dl - 375 litres of glucose standard + 125 litres of distilled watered

• 200 mg/dl glucose standard in 500 ml

added 5ml of glucose oxidase reagent, incubate for 1 hour at 37°c, and compare the extinction to the reagent blank at 540nm.

if the sample's absorbance valued was too high, dilute the previously obtained supernatant 2x with distilled watered and repeated the procedure.

URINALYSIS

MATERIAL:

- Gloves and eyeglasses are examples of personal protective equipment (plus apron if available).
- Check the expiration date on reagent strips before using.
- Container for reagent strips with colour chart.
- Urine collecting container should be clean.

METHOD:

before removing the container from the patient, wash your handed and put on gloves.

• remove the covered and thoroughly immerse the reagent stripped in the urine by removing the lid. Removed the urine container as soon as possible and tapped on the side to shook out any remaining drips.

• allow any residual urine to drain out by holding the stripped at an angle.

• wait the specified amount of time (as specified on the reagent stripped container) before comparing the findings to the colour chart on the reagent stripped container's side. Avoided touching anything, including the side of the reagent stripped container and any other surfaces.

• remove the urine and dispose of it properly.

• as directed by your clinical waste policy, dispose of the contaminated equipment (gloves, reagent stripped, and urine container, if disposable).

• take off your gloves and wash your handed.

RESULT:

Table 1: Data from blood and urine tests are displayed in a table along with age and gender

SR.NO.	AGE (MALE/FEMALE)	BLOOD				URINE	
		HbA1c%	RPG	FBS	OGTT	FBS	OGTT
1	71(F)	14.0	239	441	616	+2	+3
2	45(M)	7.4	145	152	210	+2	+3
3	72(F)	6.3	500	175	150	AB	AB
4	46(M)	6.0	225	155	206	+2	+2
5	50(M)	5.7	175	118	218	+3	+3
6	65(M)	5.7	422	71	317	AB	+2
7	45(M)	5.9	387	126	172	AB	AB
8	48(M)	7.0	232	130	213	AB	+3
9	62(M)	5.8	135	201	149	AB	AB
10	51(M)	8.5	311	213	190	AB	AB
11	57(M)	6.8	89	188	219	AB	AB
12	62(M)	8.9	426	158	241	AB	+3
13	58(M)	6.4	126	188	361	+1	+3
14	73(M)	5.9	292	117	186	AB	+2
15	64(M)	9.6	81	264	441	AB	+3
16	50(M)	6.2	256	147	193	AB	AB
17	53(F)	12.0	145	363	607	+2	+2
18	69(M)	5.7	67	109	173	AB	AB
19	63(M)	6.6	406	114	110	+2	+3
20	70(F)	6.8	128	131	120	+2	+2
21	54(M)	11.6	103	348	213	+3	+1
22	47(M)	13.7	222	359	485	+1	+2
23	58(F)	6.7	159	161	392	AB	AB
24	63(F)	5.7	146	130	170	AB	+1
25	60(F)	5.9	221	132	170	+2	+3

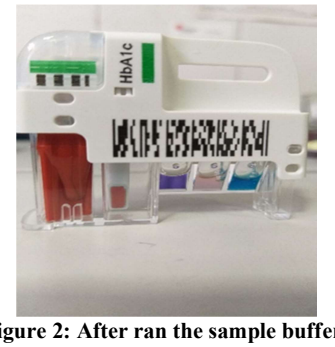

Figure 2: After ran the sample buffer reagent color got changed

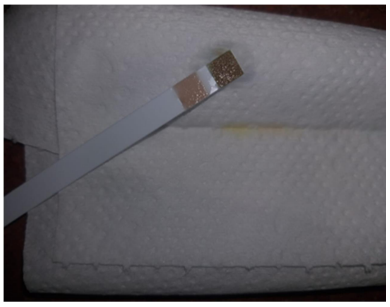

Figure 3: Stripped changed the color

DISCUSSION:

The increasing worldwide prevalence of these conditions, the identification of pre-diabetic and long latent diabetic phases, and the availability of appropriate, reliable, high-performance, and acceptable tests to aid in the detection of hyperglycaemic conditions are all reasons to screen for prediabetes and diabetes. Although no study on the impact of hyperglycaemia screening on clinical outcomes (disease and mortality) has been published, diabetes testing does not appear to have any detrimental psychological consequences. On the other hand, the impact of a "prediabetic" diagnosis remains unknown.

A1c of less than 5.7 percent is normal, 5.7 to 6.4 percent indicates prediabetes, and 6.5 percent or higher indicates diabetes. The average range for a random blood glucose test is higher than or equal to 200.

FBS and OGTT results, as well as gender, for both blood and urine tests. Males Dominate females, implying that females are inferior to males. It also demonstrates that some patients have diabetes in their blood but not in their urine, and that others have glucose in their urine but not in their blood.

Blood tests are performed in conjunction with their gender and age. Patients aged 45 to 75 are covered, according to the chart. The FBS concentration should be between 70 and 110 mg/l, whereas the OGTT concentration should be between 70 and 140 mg/l. There are two patients with normal ranges in the FBS and 23 patients with diabetes in the OGTT, whereas there are two patients with normal ranges in the FBS and 23 patients with diabetes in the OGTT.

CONCLUSION:

Diabetes mellitus is the pandemic of the century, and diabetes will continue to increase until better early detection tools are developed. This study focuses on the many forms of diabetes and the most efficient diagnostic tools and criteria for diabetes and prediabetes diagnosis. Diabetes is clearly a complicated illness with a significant number of genes involved in its progression. The accurate identification of the genetic underpinnings of diabetes might be a valuable tool for improving diabetes diagnosis, medication, and genetic counselling. Furthermore, we have a thorough understanding of the link between medical genetics and diabetes' chronic consequences.

Diabetes type 2 is caused by the body's inability to respond to insulin. Obesity and a lack of physical activity are two major risk factors. The standard test is the OGTT. Diabetes mellitus 2 can be avoided by living a healthy lifestyle and avoiding alcohol.

ACKNOWLEDGMENT

It is our privilege and honour to express our heartfelt gratitude to Parul University in Vadodara, Gujarat, for providing me with all of the necessary support and facilities, including cutting-edge infrastructure, advanced technological scientific laboratories, and everything else needed to complete this project.

REFERENCES:

[1] Diabetes mellitus: The epidemic of the century; Akram T Kharroubi and Hisham M Darwish; World J Diabetes. 2015 Jun 25; 6(6): 850–867.

[2] American Diabetes Association. Diagnosis and classification of diabetes mellitus. Diabetes Care. 2014;37 Suppl 1: S81–S90. [PubMed] [Google Scholar]

[3] Craig ME, Hattersley A, Donaghue KC. Definition, epidemiology and classification of diabetes in children and adolescents. Pediatr Diabetes. 2009;10 Suppl 12:3–12. [PubMed] [Google Scholar]

[4] Galtier F. Definition, epidemiology, risk factors. Diabetes Metab. 2010; 36:628–651. [PubMed] [Google Scholar]

[5] History of Diabetes; By Dr. Ananya Mandal, MD, Reviewed by April Cashin-Garbutt, MA (Editor)

[6] Diabetes Mellitus Screening; Advait Vasavada; Leslie Faith M. Taub; Advait Vasavada; Leslie Faith M. Taub. Affiliations, AIIMS New York University; August 27, 2021.

[7] Diabetes Mellitus; Choose the Right Test - ARUP Consult; Grace M. Kroner, PhD Clinical Chemistry Fellow at ARUP Laboratories and University of Utah, Lauren N. Pearson, DO, MPH Assistant Professor of Pathology (Clinical), University of Utah.

[8] Diabetes Tests; August 10, 2021; Centres for Disease Control and Prevention; www.cdc.gov/diabetes/basics/getting-tested.html

[9] The National Institute of Diabetes and Digestive and Kidney Diseases; Diabetes Tests & Diagnosis; American Diabetes Association. Classification and diagnosis of diabetes. Diabetes Care; December 2016.

[10] Definition and diagnosis of diabetes mellitus and intermediate hyperglycaemia Report of a WHO/IDF Consultation 2006; p.g., 32.

[11] Haemoglobin A1C (HbA1c) Test: Medline Plus Medical Test; 15-Sept-2021, review article.

[12] Urine testing for diabetic analysis; Janet Marsden and Dianne Pickering; Journal List: Community Eye Health v. 28(92); 2015.