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IDENTIFICATION OF FUNCTIONALAL GROUPS IN URINE OF DIABETES MELLITUS (DM) TYPE II PATIENTS USING INFRARED SPECTRUMETER

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ABSTRACT

A preliminary study was performed to identify functional groups in urine of diabetes mellitus (DM) type II patients. In this research, samples of DM patients were compared with non-DM samples. Sample test is done on the urine of fasting and urine 2 hours after eating, and tested its sugar level content. The test results of urine samples with IR spectrometer FTIR Type MB3000 ABB in Solid Physics Laboratory showed that, DM patients were identified on samples Fasting urine was alcohol functional group (OH stretch, 3200 -3650 cm-1), functional group Alkene (CH stretch, 2800-2900 cm-1), Carbon Dioxide functional groups (C≡C and C≡N, 2350 cm-1), and Amide functional groups (CO Stretch, 1650-1690 cm-1). Fasting sugar level 239 ml /dl. For the urine Samples 2 hours after meals, the functional groups of Alcohol (OH stretch, 3200-3650 cm-1), Alkene functional groups (CH Stretch, 2850-3000), Carbon Dioxide functional groups (C≡C and C≡N, 2350 cm-1), Alkene functional groups (C≡C, 2100-2210 cm-1), Amide functional groups (C = O, 1640-1670 cm-1), Alkene functional groups (CH bend, 1450-1470 cm- 1), and the functional group of Esters, (CO Stretch, 1000-1300 cm-1). And sugar levels 2 hours after eating 408 ml / dl. In non-DM samples, the samples of Fasting Urine (blood sugar level 100 ml / dl) and urine 2 hours after meal (blood sugar level 140 ml / dl), did not show significant differences, tended to be same functional and absorbance, Alcohol functional groups (OH stretch, 3200-3650 cm -1), Carbon Dioxide functional groups (C≡C and C≡N, 2350 cm-1), and Amide

functional groups (C = O, 1640-1670 Cm-1). Significant differences in both DM and non-DM samples were identified in the presence of an alkene-functional group (CH Stretch, 2850-3000), Alkene functional groups (C≡C, 2100-2210 cm-1), Alkene functional groups (CH bend, 1450 -1470 cm-1), and the functional group of Esters, (CO Stretch, 1000-1300 cm- 1). Other things quite different on the Amide functional (C = O, 1640-1670 cm-1), although appearing in both samples, the highest absorbance values are in urine samples during DM patients, compared with DM.

INTRODUCTION

Diabetes Mellitus (DM), according to WHO is a chronic disease that occurs either when the pancreas does not produce enough insulin or when the body cannot effectively use the insulin produced (WHO, 2008). Diabetes erupts are a group of disorders characterized by elevated blood sugar levels. Diabetes erupts is a state of chronic hyperglycemia accompanied by various metabolic abnormalities due to hormonal disturbances, which lead to various chronic complications in the eyes, kidneys, nerves and blood vessels.

Someone can be said DM when diagnosed with DM diagnostic criteria and impaired glucose tolerance i.e. blood glucose level (venous plasma) ≥ 200 mg / dl, fasting blood glucose (venous plasma) ≥ 126 mg / dl, plasma glucose level ≥ 200 mg / Dl at 2 hours after glucose load 75 gram on Oral Glucose Tolerance Test (TTGO) (PERKENI, 2011).

Table 1. Diagnostic Criteria For Diabetes And Intermediate Hyperglycemia

Diabetes

Fasting plasma glucose 2–h plasma glucose*	≥7.0mmol/l (126mg/dl) or ≥11.1mmol/l (200mg/dl)

Impaired Glucose Tolerance (IGT)

Fasting plasma glucose 2–h plasma glucose*	<7.0mmol/l (126mg/dl) and ≥7.8 and <11.1mmol/l (140mg/dl and 200mg/dl)

Impaired Fasting Glucose (IFG)

Fasting plasma glucose 2–h plasma glucose*	6.1 to 6.9mmol/l (110mg/dl to 125mg/dl) and (if measured) <7.8mmol/l (140mg/dl)

Diabetes type II is a common form of diabetes mellitus. Without proper diagnosis and treatment, this disease can cause serious health problems. If you have type 2 diabetes, also known as adult-onset diabetes or diabetes that does not depend on insulin, the body You have enough insulin but are not able to use it properly. This disease is different from type 1 diabetes, where the pancreas cannot produce insulin. In type 2 diabetes, the pancreas actually works normally but somehow the cells in the body cannot use glucose inside Blood as a source of energy. And over time, high glucose in your blood will harm the body. About 90 to 95% of diabetic patients are diagnosed with type 2 diabetes. This disease often strikes in adulthood, by age 40 or older.

This study will examine functional groups in urine of type 2 diabetes mellitus compared with non-DM. The technique used to examine functional groups is infrared spectroscopy. Spectroscopy is the study of matter and its attributes based on light, sound and particles emitted, absorbed or reflected by the material. In the history record spectroscopy refers to the branch of science where "visible light" is used in material structure theories as well as qualitative and quantitative analysis. In modern times the definition of spectroscopy develops as new techniques are developed to exploit not only visible light, but also other forms of electromagnetic and no electromagnetic radiation such as microwaves, radio waves, electrons, phonons, sound waves, X-rays and so on. FTIR spectroscopy (Fourier transform infrared) is one of the best analytical techniques in the process of identifying the molecular structure of a compound. The main component of FTIR spectroscopy is the Michelson interferometer which has the function of disentangling (dispersing) infrared radiation into frequency components Spectrum produced from spectroscopy is the unique or spectrum of a material having different shapes for other compounds. Basically FTIR (Fourier Transform Infra Red) spectroscopy is the same as IR disperse spectroscopy, which distinguishes it from the development of the trigger before the infrared light.

METHODS

The use of the Michelson interferometer provides an advantage over the FTIR method compared to conventional infrared spectroscopy methods as well as other spectroscopic methods. Among them is the information molecular structure can be obtained precisely and accurately (has a high resolution). Another advantage of this method is that it can be used to identify

samples in different phases (gas, solid or liquid) . The difficulties encountered in the identification with FTIR spectroscopy can be supported by data obtained using other spectroscopic methods.

Infrared spectroscopy methods can be used to identify an unknown compound, because the resulting spectrum is specific to the compound.

Qualitative analysis with IR. As a complement to obtain structural information from the compound through interpretation. IR spectra can be used IR correlation table (Table 2) which contains information where the functional group absorbs It is generally useful to classify all regions into three to four wide areas. One way is to categorize some near IR areas (0.7-2.5 μ); Fundamental areas (2.5-5.0 μ); And far IR region (50-500 μ).

Table 2. Typical uptake of some functional groups of IR

Fungsional group	Type of vibration	Characteristic Absorptions (cm-1)	Intensity
Alcohol			
O-H	(stretch, H-bonded)	3200-3600	Strong, broad
O-H	(stretch, free)	3500-3700	
Alkane			
C-H	Stretch	2850-3000	
-C-H	Bending	1350-1480	Variable
Alkene			
=C-H	Stretch	3010-3100	Medium
=C-H	Bending	675-1000	Strong
C=C	Stretch	1620-1680	Variable
Alkyl Halide			
C-F	Stretch	1000-1400	Strong
C-C1	Stretch	600-800	Strong
C-Br	Stretch	500-600	Strong
C-1	Stretch	500	Strong
Alkyne			
C-H	Stretch	3300	Strong, sharp
-C≡C-	Stretch	2100-2260	Variable, not present asymmetrical alkynes
Amine			
N-H	Stretch	3300-3500	Medium (primary amines have two bands; secondary have one band, often very weak)

Fungsional group	Type of vibration	Characteristic Absorptions (cm-1)	Intensity
C-N	Stretch	1080-1360	Medium-weak
N-H	Bending	1600	Medium
Aromatic			
C-H	Stretch	3000-3100	Medium
C=H	Stretch	1400-1600	Medium-weak, multiple bands

Analysis of C-H out-of-plane bending can often distinguish substitution patterns

Carbonyl	Detailed information on carbonyl IR		
C=O	Stretch	1670-1820	Strong
(conjugation moves absorptions to lower wave numbers)			
Ether			
C-O	Stretch	1000-1300 (1070-1150)	Strong
Nitrile			
CN	Stretch	2210-2260	Medium
Nitro			
N-O	Stretch	1515-1560 & 1345- 1385	Strong, two bands

The absorption of infrared radiation causes an energy change (ΔE) and is expressed as:

ΔE =h υ, with vibration levels. Where h = Planck constant (6.6242 x erg det) and v=frequency constant in Hertz (Hz). The relationship between frequency and wavelength (λ) is expressed as: $v = \frac{c}{\lambda}$ Where c is speed of light (3 x 10^{10}cm dt^{-1}) and λ is expressed in cm The opposite wavelength

(ṽ) represents the number of waves per cm. So, $\tilde{v} = \frac{c}{\lambda}$

The number of waves (ṽ) is directly proportional to the frequency or energy, therefore the horizontal portion of the infrared spectrum is usually expressed as the number of waves (ṽ) in cm^{-1}. So the wave Numbers are directly proportional to the Frequency, $V = c. \tilde{v}$

The vibrational frequencies of the bonds must be reasonably precise, as they are, calculating the vibrational frequencies of the spheres and the spring system. Hooke's law can help to estimate the area where vibrations

$$\overline{\upsilon} = \frac{1}{2\prod C}\left(\frac{k}{\frac{m1 + m2}{m1 X m2}}\right)1^{1/2}$$

Based on Hooke's Law:

υ = number of waves (cm^{-1}, m1= Atomic mass 1 (g), m2= Atomic mass 2 (g)
K = Styling force for bond (g / s)

The formula used to calculate the amount of energy absorbed:
$E = h.\nu = h.C / \lambda = h.c / v$

The equipment used in this research is FTIR spectrum (Fourier Transform Infrared) MB 3000 type used to identify mineral contents. Material used in this research is Window Kbr. Sample used, among others, fasting water with diabetes mellitus, urine after eating DM, fasting urine is non=DM, and urine after eating is non-DM

RESULTS

The diagnosis of DM should be based on examination of blood glucose levels. Patients with very high sugar content then the sugar will be removed through the urine. Lab test results for blood glucose levels of DM patients stated, when fasting of 239 ml / dl and after 2 hours of eating is 408 ml / dl. This result is paired with the results of a fasting urine sample test and after 2 hours of feeding with IR spectroscopy method. Based on the results of research on the IR spectrum obtained from urine samples in patients with DM disease.
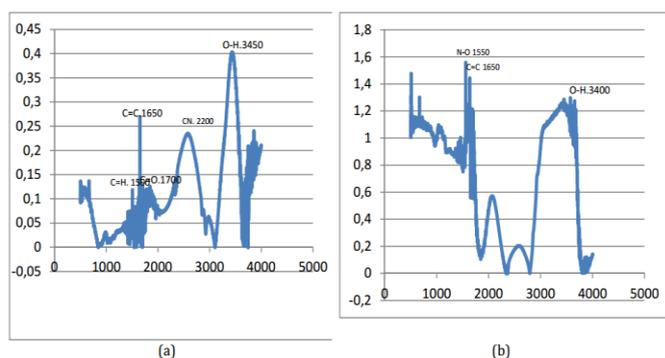

Figure 1. Spectra (a) sample Fasting urine DM, (b) Urine sample 2 hours after meals

In the above illustration, for samples (a) Fasting urine is an alcohol functioning group (OH stretch, 3200-3650 cm-1), an alkene functional group (CH stretch, 2800-2900 cm- 1), Carbon Dioxide functional group (C≡C and C≡N, 2350 cm-1), and the Amide functional group (CO Stretch, 1650-1690 cm-1).

For samples (b), the functional groups of Alcohol (OH stretch, 3200-3650 cm-1), Alkene functional groups (CH Stretch, 2850-3000), Carbon Dioxide functional groups (C≡C and C≡N, 2350 cm- 1), Alkene functional groups (C≡C, 2100-2210 cm-1), Amide functional groups (C = O, 1640-1670 cm-1), Alkene functional groups (CH bend, 1450- 1470 cm-1), And the functional group of Esters, (CO Stretch, 1000-1300 cm-1). The results of blood glucose and IR spectrum examination showed significant changes. Blood glucose levels of DM patients after 2 hours of feeding were very high, when compared to the IR spectra of the two samples there were differences in the identified functional groups, as well as the absorption rate of each functional group. Very prominent is the OH group of samples after 2 hours of eating that has a high uptake, a wide peak. In addition, there were also identified new functional groups namely, C-O, and C-H groups.

For the results of Non-DM samples, lab test results for blood glucose levels, when fasting 100 ml / dl and after 2 hours of meal at 140 ml / dl. And the lab test results of urine samples during fasting and after 2 hours of feeding with IR are shown in the figure. 2 follows.
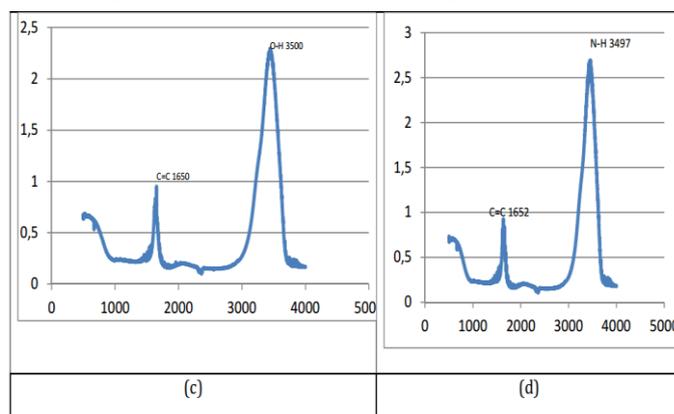

Figure 2. IR spectrum (c) Non-DM urine fasting, (b) Non-DM urine after 2 hours of feeding

In the Non-DM sample, the identified functional groups showed the same tendency, however, there were significant differences in absorbance, i.e. in the Alcohol function (OH stretch, 3200-3650 cm-1), Carbon Dioxide functional groups (C≡C and C≡N, 2350 cm-1), and the Amide functional group (C = O, 1640-1670 cm-1). Significant differences in both DM and non-DM samples were identified in the presence of an alkene-functional group (CH Stretch, 2850-3000), Alkene functional groups (C≡C, 2100-2210 cm-1), Alkene functional groups (CH bend, 1450 -1470 cm-1), and the functional group of Esters, (CO Stretch, 1000-1300 cm-1). Other

things quite different on the Amide function (C = O, 1640-1670 cm-1), although appearing in both samples, the highest absorbance values are in the urine sample during DM patients, compared with fasting DM urine and Non-DM samples. OH stretch group in hydrogen group OH bonds is seen in the frequency characteristics of 3300 cm-1 which is the spectrum region of Glucose compound. And the C-C and C-O bonds on the spectrum show a fairly close absorption band between 1100-1000 cm-1 in the spectrum of carbohydrate compounds.

Factor causes of the rise in blood glucose levels are simple carbohydrates that have chemical bonds only one and easily absorbed into the bloodstream so that it can directly raise blood sugar levels. Complex carbohydrates are carbohydrates that are difficult to digest by the intestines.

The absorption of these complex carbohydrates is relatively slow, providing a longer satiety and not rapidly raising the blood sugar levels in the body. Complex carbohydrates are converted into glucose longer than simple carbohydrates so it is not easy to raise blood sugar levels and better provide energy that can be used in stages throughout the day. Carbohydrates that are not easily broken down into glucose are found in nuts, fiber (vegetables and fruits), starches, and tubers. Because of it, its absorption is slower so as to prevent the increase of blood sugar level drastically. Conversely, easily absorbed carbohydrates, such as sugar (whether sugar, red sugar or syrup), grain products (bread, pasta) will actually accelerate the increase in blood sugar (Susanto, 2013).

Metabolism of carbohydrates and diabetes mellitus are two links that cannot be separated. The link between carbohydrate metabolism and diabetes mellitus is explained by the presence of insulin hormone. Diabetes mellitus suffered damage in production and insulin working system, while in indispensable in regulating carbohydrate metabolism. As a result, people with diabetes mellitus will experience interference with carbohydrate metabolism.

CONCLUSION

The results stated that blood glucose levels of DM patients increased significantly after 2 hours of eating. This is seen in the results of laboratory tests and IR spectrum, where functional groups indicating elevated glucose levels of urine are the OH, C-C and C-O groups.

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