FOLLOW UP THE CONTROL DIABETES MELLITUS CASES BY HBA1C METHOD AMONG PATIENTS ATTENDING AL-NASSIRYAH DIABETIC CENTER

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ABSTRACT
This paper reports the results of study carried out in Al – Nassiriya city in the center of diabetic and endocrine glands from the period of 1.7.2008 -30.8.2008. It aims at quantifying HbA1c ratio in diabetic patient from type 1 and type 2 to show the well or poorly control diabetes mellitus.in this study has been collected 50 patient randomly (male 21 and 29 female), in different age from 7-65 years old.
Result :The percent of patients with Glibenglamide drug was 24% same percent with repaglinide ,but the repaglinide improve glyceamic control without causing weight gain or sever hypoglycemia and provides complete control by effectively reducing fasting blood glucose and HbA1c compared to glibenglamide ,metformin and insulin.The percent with Glibenglamide drug taken with metformin it was 27%,the same percent in patient with insuline drug. Conclusion :controlled diabetes ,not much glycosylated hemoglobin represented in a vocational diploma or college degree constituted of cases ,while uncontrolled diabetes ,much more glycosylated hemoglobin represented in illiterate case.

INTRODUCTION
Diabetes is not a single disease, it is a heterogeneous group of syndromes all characterized by an elevation of blood glucose caused by relative or absolute deficiency of insulin. Insulin is the peptide hormone that secreted from cells located in the islets of langerhans in pancreas (B-cell produce insulin) this hormone play an important role in regulating the metabolic activities of the body (help maintain the homeostasis of blood glucose).
Diabetics can be divided into two groups based on their requirements for insulin: 1- IDDM or type 1 (insulin dependent diabetes mellitus). 2- NIDDM or type 2(non insulin dependent diabetes mellitus).
Type 1:-
Is characterized by an absolute deficiency of insulin caused by massive B-cell lesion or necrosis. Loss of B-cell function may be due to invasion by viruses, action of chemical toxins or usually through the actions of autoimmune antibodies directed against the B-cell. The type (1) diabetic shows classic symptoms of insulin deficiency (polydipisia, polyphagia and polyuria). The development and progression of neuropathy, nephropathy and retinopathy are directly related to the extent of glycemic control (most often measured as blood levels of HbA1C).
The age of onset in type (1) usually during childhood or puberty. The treatment of type (1) diabetes: - the purpose in treating type (1) diabetes is to control hyperglycemia by (injected) insulin and maintain acceptable levels of HbA1c and avoid ketoacidosis.
Insulin therapy: - all patients with type (1) require treatment with insulin. There are 3 type of insulin available from different species: beef, pork and human

\*Iraq-al Nassiryah city –the center of diabetic and endocrine glands
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(is produce by special strain of E. coli through recombinant DNA technology).

The insulin preparations: - the onset of action, peak effect and duration of action are determined both by insulin type and by the physical and chemical form of the insulin which are :-

1- Neutral insulin
2- Isophane insulin
3- Lent insulin

Type2:-
Is much commoner than type (1), occurs in patients over age 35 years (it is increase with age and with increasing obesity) genetic factor rather than or autoimmune antibodies are apparently causal. In some cases, insulin resistance is due to a decrease number or mutation of the insulin receptor .undefined defect viruses in the events that occur after insulin binds to its receptor is believed to account for resistance in most patient. The metabolic alterations observed are milder than those described for IDDM, but the long term clinical consequences can be just as devastating (for example :- vascular complications and subsequent infection can lead to amputation of the lower limbs).

Treatment of type (2) diabetes: - the purpose in treating is to maintain blood glucose concentrations within normal limits and to prevent the development of long term complication of the diseases.

Wight reduction, exercise and dietary modification decreases insulin resistance and correct the hyperglycemia (2) diabetes in some patients. (1) insulin therapy may be requiring to achieve satisfactory serum glucose levels.

Oral hypoglycemic agents :- these should not be given to type (1)

(A) sulfonylurea :-
This class of drug rely on the ability of pancreas to secret insulin , reduction of serum glucagons levels and increase binding of insulin to target tissue and receptors . several receptors have suggest that sulfonylurea may reduce platelet aggregation , it is likely that the anti platelet affect are secondary to the improvement in blood glucose concentrations produced by these drugs.

for example :- glibenclamide(secondary generation ), daily dose range (2.5-15mg)/day .it is give alone or in combination with other oral hypoglycemic agents .

(B) biguanides:-
metformin (trade name glucophage) is the only biguanide available in the uk. May be used alone or in combination with the sulfonylurea or repaglinide or insulin . it is acting primarily by decreasing hepatic glucose out put , largely by inhibiting gluconeogenesis .it’s differs from the sulfonylureas in not stimulating insulin secretion.

Glucophage is a viable in 500mg, 850mg and 1000mg. daily dose 1-3g/day.

( C ) Repaglinide (trade name novo norm)(0.5 , 1.0 , 2.0 mg) :- it is the first in a new class of drugs , the meglitinids, to be marketed in the uk. Repaglinide not sulfonyl urea , it has actions in common with this group of drugs. It binds to ATP-sensitive potassium channels of the pancreatic B-cells causing the release of insulin. It is take three time in day before meals (half life 2hr ). It may be used in combination with the metformin to produce a synergistic effect. It has shown too lower HbA1C level. Novonorm improves glycemic control with out causing weight gain or sever hypoglycemia . Repaglinide provides triple control :-
1-fasting blood glucose reduce by 74 mg/dl.
2-postprandial blood glucose reduce by 115 mg/dl.
3-HbA1C reduce by 2.7%.

It is reducing the risk of sever hypoglycemia by 60% compared with sulphonylurea and it will suited to patient with renal impairment due to its hepatic metabolism and elimination .

The maximum recommended dose is 16 mg/day

If HbA1C < 8 % 1 mg before each meal

If It is HbA1C > 8 % 2 mg before each meal
Glycated Hemoglobin
HbA1c, glycated hemoglobin, is an indicator of long-term glycemic control. In Adults, hemoglobin are a mixture of three forms: HbA1, HbA2, and HbF, with HbA1 predominating. Hemoglobin A1 consists of three subforms: Hb A1a, Hb A1b, and Hb A1c with Hb A1c predominating. The term glycated hemoglobin describes a chemically stable conjugate of any of the forms of hemoglobin with glucose. Glycated forms of hemoglobin are formed slowly, nonenzymatically, and irreversibly at a rate that is proportional to the concentration of glucose in the blood.

The level of glycated Hemoglobin in a blood sample provides a glycemic history of hemoglobin glycation over the life span of the erythrocyte, the cell that contains the hemoglobin. The average lifespan of the erythrocyte is 120 days, and glycated hemoglobin describes the average glucose levels in the blood over that life span.

The Specimen
The preferred specimen is whole blood that has been collected in EDTA, heparin, or fluoride anticoagulant. Capillary blood may be used for some procedures such as immunoassay. A hemolysate of washed red blood cells is tested. Whole blood may be stored for 1 week at 4_C. Hemolystate may be stored for 4 to 7 days at 4_C or 30 days at –70_C.

MATERIALS & METHODS
1-groups of patients the study were included 50 patients with history of diabetes mellitus from private AL-Iraq medical lab. In Nassiriya city. All patients were examined by the quantitative colorimetric determination of glycohemoglobin in whole blood in private AL-Iraq medical lab.
2-Collection of blood samples(2.5ml) of blood were collected From patients with history of diabetes mellitus . The sample then put in a tube with EDTA , allowed at room temperature for about 5 min.
The chemical method were used for the purpose of checking the collected blood samples.

**HbA1c Test**

**Principle:**

Using stanbio glycohemoglobin (pre-fil) Procedure No.P350.

This kit is quantitative colorimetric determination of glycohemoglobin in whole blood. In the method presented, a preparation of hemolyzed whole blood is mixed with a weakly binding cation – exchange resin. The non-glycosylated hemoglobin (HbA0) binds to the resin, leaving (HbA1) free to be removed by means of a resin separator in the supernate. The percent of HbA1 is determined by measuring the absorbance values at 415 nm of the HbA fraction and of the total Hb fraction. Calculating the ratio of absorbances (R), and comparing this ratio to that of a glycohemoglobin standard carried through the same procedure.

Results are express as HbA1 but can be converted or derived as HbA1C by using a conversion factor or when using a HbA1c value for the standard.

**Reagents:**

1- Glycohemoglobin ion-exchange resin . cat No.P351-(tubes): each tube contains 3.0ml cation – exchange resin , 8 mg/dl , buffered at PH6.9.

2- Glycohemoglobin lysing reagent, cat.No.0352 contains potassium cyanide .10mmol/l and surfactants.

3- Glycohemoglobin standard (lyophilized ), cat.No.0353-(1vial) prepared from packed human erythrocytes.

**Materials required but not provided:**

A spectrophotometer or filter photometer capable of absorbance reading at 415nm(415-420nm). Pipettors : 5.0ml, 1.0ml ,0.1ml,0.02ml Cuvettes : Hematology rocker (optional).

**Procedure**

Hemolysate preparation

1- pipette 0.1ml of each well-mixed blood sample into appropriately labeled tube and mix.

2- allow to stand for 5 minutes at room temperature (15-30C) to complete hemolysis.

3- Label pre-fil resin tubes standard (S), Unknown(U) and control (C).

4- Pipette 0.1ml of the prepared hemolysate into appropriately labeled resin tube.

5- Position a resin separator in the pre-fil tube so rubber sleeve is approximately 1-2 cm above liquid level.

6- Mix tubes on a hematology rocker

For 5 minutes . alternatively, tubes may be mixed by hand if held above the resin.

7- at the end of mixing , push resin separator into tube until resin is firmly packed in bottom of the 13mm tube.

8- Pour each supernate directly into separate cuvettes for absorbance measurements.

9- Read absorbance (Agly) of standard, unknown and control vs water at 415nm within 60minutes.

10- Pipette 5.0ml deionized water into tubes labeled standard (s),Unknown(u) and control(c).

11- Pipette 0.02ml of hemolysate into appropriately labeled tube, mix well and transfer to cuvette for absorbance reading.

12- Read absorbance (Atotal) of standard, unknown and control vs water at 415nm within 60 minutes.

**Calculation:**

For each standard and unknown calculate the ratio(R) of the glycohemoglobin absorbance as follows:

\[ R = \frac{Agly}{Atotal} \]

\[ Glycohemoglobin(\%) = \frac{R(u)}{R(s)} \times \text{concentration glycohemoglobin standard(\%)} \]

- If we use concentration of (S) is (10) the result will be of (HbA1), and if it is(7,6) it of (HbA1c)

- We can calculate the mean blood glucose estimate by following:

\[ MBG(\text{mg/dl}) = 36.7 \times \text{HbA1\%} - 185 \]
RESULTS & DISCUSSION

1- description of the study sample:
the results presented in this chapter were based on the analysis of data from a sample of 50 patient with diabetes mellitus who were referred to the lab for test of blood sugar.

The age of patients as follow:
Group (1) it was less than 20 years old.
Group (2) it was 21-35 years old.
Group (3) it was 36-50 years old.
Group (4) it was 51-65 years old.
Group (5) it was >65 years old.

all the subjects were under the therapy of diabetic drugs.

About more than (75%) (37 in number) of study sample were from Urban. A history of diabetes mellitus in family was reported in (64%) of cases(32 in number). about a quarter of study sample was illiterate(27.5%) (14 in number),while those with a (10%) of cases(5 in number) vocational diploma or college degree constituted.

The percent of patients with Glibenglamlide drug was (24%) (12 patient) same percent with Repaglininde, the percent with Glibenglamide taken with Metformin it was (27%) (13 patient), the same percent in patients with Insulin drug.

2- the relationship between drugs and the mean of HbA1c:

as shown in table (1) and figure (2), the lower mean of the HbA1c it was with the patients were taken the novonorm therapy (7,18%),while the higher mean Of HbA1c it was of the Patients were taken the daonil.

In the table( 2) and figure (3) we show the relationship the mean of HbA1c and the age of the patients and we notice that the age between 21-35 it was have lower in the HbA1c.

As the show in table(3) and figure(4) relationship between the mean of MBG and the type of the drugs , in this table we find that the lower mean MBG with the novonorm drug.

CONCLUSION

1- We find that the patients that take Novonorm drug, they have a good control on blood sugar (low HbA1C percent).

2-Glycated hemoglobin levels do not require a fasting specimen and can be used In conjunction with the patient’s self-monitored blood glucose records to determine glycemic control over the past few months.

3-It is not practical or necessary for diabetic patients to fast for 8 to 10 hours prior to a visit to a diabetes center or physician’s office for Follow-up.

ADVISE

1- It is important for diabetic patients to eat meals on a regular basis and balance their Food intake with activity versus their blood glucose level.

2- Laboratory personnel may need to communicate This information with nursing or clinic staff when initially setting up services for those patients.

3- Diabetic patients they must be make the test (HbA1C) level every 3 months to recognize the control or non control on blood sugar.

Table (1) the relationship between drug and the mean of HbA1c.

<table>
<thead>
<tr>
<th>Type of drug</th>
<th>HbA1c mean</th>
<th>HbA1c max.</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>daonil.</td>
<td>7.82</td>
<td>10.50</td>
<td>9.0</td>
</tr>
<tr>
<td>Daonil+glucophage</td>
<td>5.80</td>
<td>11.67</td>
<td>8.4</td>
</tr>
<tr>
<td>Insuli.</td>
<td>6.73</td>
<td>11.34</td>
<td>9.16</td>
</tr>
<tr>
<td>Novonorm</td>
<td>5.80</td>
<td>8.57</td>
<td>7.11</td>
</tr>
</tbody>
</table>
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Table (2) the relationship between the age and the minimum and maximum of HbA1c.

<table>
<thead>
<tr>
<th>Age</th>
<th>number</th>
<th>HbA1c man.</th>
<th>HbA1c max.</th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;20</td>
<td>7</td>
<td>6.14</td>
<td>10.8</td>
<td>9.0</td>
</tr>
<tr>
<td>20-35</td>
<td>12</td>
<td>5.80</td>
<td>9.83</td>
<td>7.6</td>
</tr>
<tr>
<td>36-50</td>
<td>18</td>
<td>7.65</td>
<td>11.67</td>
<td>10.3</td>
</tr>
<tr>
<td>51-65</td>
<td>5</td>
<td>6.64</td>
<td>11.34</td>
<td>9.33</td>
</tr>
<tr>
<td>&gt;65</td>
<td>8</td>
<td>6.81</td>
<td>8.99</td>
<td>10.55</td>
</tr>
</tbody>
</table>

Table (3) the relationship between the drug and the MBG mean

<table>
<thead>
<tr>
<th>Drug</th>
<th>MBG Man</th>
<th>MBG Max</th>
<th>mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Daonil</td>
<td>189</td>
<td>309</td>
<td>233</td>
</tr>
<tr>
<td>D+G</td>
<td>101</td>
<td>389</td>
<td>248</td>
</tr>
<tr>
<td>Insulin</td>
<td>142</td>
<td>365</td>
<td>218</td>
</tr>
<tr>
<td>Novonorm</td>
<td>101</td>
<td>222</td>
<td>111</td>
</tr>
</tbody>
</table>

Figure (2) the relationship between drug and the mean of HbA1c.
Figure (3) the relationship between the HbA1c mean and the ages of patients

Figure (4) the relationship between the MBG mean and the type of drugs
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REFERENCES

HbA1C

متتابعة مرضى السيطرة على مرض داء السكري بواسطة HbA1C

المراجع:

د. عماد أحمد عبد، د. عبير هادي بدر

الخلاصة:

أجريت هذه الدراسة في مدينة الناصرية في مركز السكر والغدد الصماء التخصصي من تاريخ ٢٠٠٨-٧-٢٠٠٨ ونهاية ٨-٧-٢٠٠٨.

الهدف منها حساب نسبة HbA1C عند مرضى السكر من النوع الأول والثاني لمتابعة السكر المسيطر عليه من السكر الغير مسيطر عليه في هذه الدراسة تم جمع ٥٠ مريض عشواً بينهم (١٢ دكماً و٨ بانياً) تتراوح أعمارهم بين ٥-7 عام.

النتائج: نسبة المرضى الذين يتولون علاج الدانونوغي ٤٢% كنسبة النفوفونوم لكن النفوفونوم بطور من السيطرة على السكر بدون ان يسبب زيادة في الوزن او بعوادحس بالسمن من خلال تقليله بالمقارنة مع HbA1C و FBS و上がون علاج الدانونوغي والانسولين فينتيرد، نسبة الذين يأخذون علاج الدانونوغي والانسولين ٢٣% كنسبة الدين يأخذون الانسولين.

الاستنتاج: يوجد في هذه الدراسة ان السكر المسيطر عليه يتمثل في دوي المستوى الثقافي العالي في حين ان السكر الغير مسيطر عليه يتمثل في دوي المستوى الثقافي البسيط.

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