Plasma Fructosamine as Indicator of Past Glycaemic Control in Non-Insulin Dependent Diabetes Mellitus (NIDDM)

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Fructosamines are glycated plasma proteins which predict retrospective glycaemia of the past 2-3 weeks; while glycated haemoglobin (HbA_{1C}) foretells the same for the past 8 – 10 weeks. The glycaemic levels in 30 female patients of Non-Insulin. Dependent Diabetes Mellitus (NIDDM) having mean age of 47 years were assessed by measuring fructosamine levels in plasma and HbA_{1C} in the blood. A highly significant correlation was found between plasma fructosamine level and HbA_{1C} r=0.46, P<0.01. It was found that fructosamine estimation is an equally valuable predictor of glycaemic control in terms of reliability and precision. Moreover, it is comparatively simple and economical to perform than to estimate HbA_{1C}.

Key words.Fructosamine, HbA_{1C}, NIDDM

The most common form of diabetes seen in Pakistan is Non-Insulin Dependent Diabetes Mellitus (NIDDM). It's prevalence in all diabetics varies from 84.5% to 94.3%[1]. The effective management of diabetic patients requires knowledge of average glycaemia prevailing in the recent past[2]. Fasting plasma glucose test (FPG) is used but it is difficult to fast for every subject[3]. It is also affected by patient's compliance prior to test[4]. Its levels fluctuates widely so a single measurement can be extremely misleading to assess the state of glycaemia[5]. An other parameter to estimate glycaemia is to assess level of glycated haemoglobin (HbA_{1C}). It is quantitative index of glycaemia for the period of 6-10 weeks prior to venepuncture due to long half life of erythrocyte that is 60 days[6]. But HbA_{1C} is expensive and time consuming to perform. A recent method of assessing the past glycaemia is measurement of fructosamine level Non-enzymatically glycated serum proteins are known as fructosamine and it's estimation provides an indicator of blood glucose level in previous 2–3 weeks[7]. It is due to shorter half life of plasma proteins especially albumin, the average is 17 days[8] . Based upon reducing ability of fructosamine Johnson et al., in 1982 introduced a rapid colorimetery method termed as fructosamine assay. Fructosamine reduces the dye nitroblue tetrazolium (NTB_4) in alkaline pH. Measurement of this process is done by noting the subsequent change in the optical density[9]. Fructosamine assay appears to offer many advantages in terms of precision, cost, time & ease of analysis[10].

Aims and objectives

This study was designed to assess the correlation of fructosamine and HbA_{1C} in estimating glycaemic control in NIDDM patients.

It is yet to be decided whether fructosamine can replace HbA_{1C} as an index of glycaemic control in NIDDM patients or it is a useful adjunct to HbA_{1C} in estimation of past glycaemic control.

Patients and methods

Thirty female patients of NIDDM attending the Diabetic Clinic of PMRC at Sir Ganga Ram Hospital for monitoring their glycaemic control were included in this study at random. The age range was 35-60 years. All patients were free of diabetic complications. They were having normal liver and renal functions and having duration of diabetes less than 5 years. Venous blood was taken into EDTA tubes, was stored at 2-8°C to measure HbA_{1c} within a week. Plasma was separated and stored at 2-8°C to perform test on fructosamine for 2 weeks.

HbA_{1c} estimation by Blick and Liles[11].

1. HbA_{1c} estimation was done by Standard Kit method (Biosystem). This test is based upon short column chromatography. Cation exchange resin was provided in disposable columns. Depending upon the specific cationic nature of HbA_{1c} , it is eluted separately by using a suitable buffer. The absorbance was noted at 415 nm. The results were calibrated by using control containing freezed dried erythrocytes. The control was made to run with each set of samples.
2. Fructosamine assay by Jhonson et al[9]. Fructosamine assay by Kit method (ROCHE). After adding the reagent blank, standard and sample were incubated at 37°C. ΔA (A_2-A_1) was calculated by noting the change in optical densities at 10 min (A_1) and at 15 min (A_2) of incubation (Absorbance at 550 nm). For calibration of results lyophilized human serum with known range of glycosylation (Precipath) was used.
3. Total serum protein estimation was done by Biuret[12] method (Kit by Randox).

Results

In total 30 patients of NIDDM, the HbA_{1C} and serum fructosamine assay were performed. The values of results are as follows:

Table 1: Age, HbA_{1c}, Fructosamine, and serum total protein , in NIDDM patients.

Parameters	Mean ±SD	Range
Age (years)	47.30±7.98	35.00-60.00
HbA_{1c} (%)	10.82±2.46	5.20-13.50
Fructosamine (µmol/L)	387.93±94.92	250.00-631.00
Serum Total Protein (*g/dL*)	7.31±0.63	6.60-8.4*

*The total serum proteins were within the normal range.

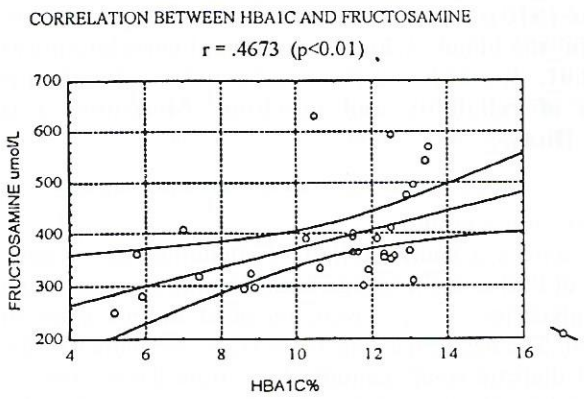

Figure 1: Shows the linear correlation analysis of Fructosamine and HbA_{1c}.

Solid line in the graph indicates the positive linear correlation between Fructosamine and HbA_{1C} . Broken lines above and below solid line indicate +1SD and -1SD of regression from the mean.

The fructosamine level correlated well with HbA_{1C} and this correlation was found to be highly significant r=(0.467, P<0.01). There is some discordance in the values of two parameters in some patients. Discrepancy of the type when a patient has high HbA_{1c} value with comparatively less fructosamine concentration indicates worse control of blood sugar in past that is two months back. Conversely a low HbA_{1c} value with high fructosamine concentration in the same patient states the recent deterioration of glycaemic control. Furthermore, no significant correlation was found between serum fructosamine and total serum proteins while the later were within the normal range.

Interpretation of results

The results of this study can be analyzed in a better way if the NIDDM patients are divided according to their glycaemic control into different categories according to Hom[13]

Table 2: Comparison of HbA_{1C} and Fructosamine in terms of glycaemic control categories

Parameters	n=	Good	Moderate	Poor	Very Poor
HbA_{1C} %Normal range 4.2 – 6.2%	30	5–8 (5)*	8.1–11 (6)	11.1–13 (15)	>13.0 (4)
Fructosamine' Normal range 208 – 280 µmol/L	30	250–300 (4)	301–350 (6)	351–450 (14)	>450 (6)

*The figures in parenthesis denote the number of patients.

Comparison of Glycaemic Control Categories

It was found that almost 50% of the total patients were having poor glycaemic control. Both parameters almost equally detect the good, moderate and poor control equally. There is minor discordance among different categories. However greater number of patients in last category indicates that fructosamine assay is more sensitive parameter to detect the very poor control.

Our results are still better expressed by the following frequency distribution curves for each parameter.

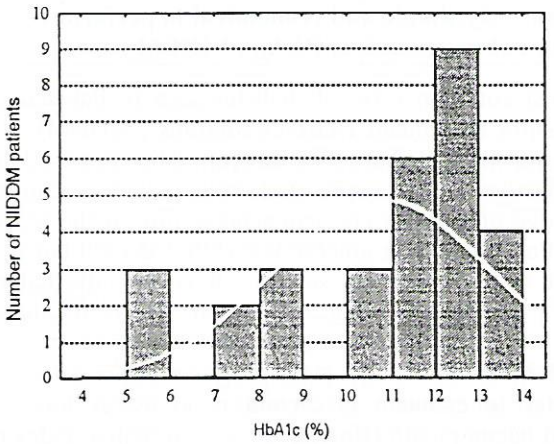

Fig 2: Frequency distribution of NIDDM patients in HbA_{1c} test

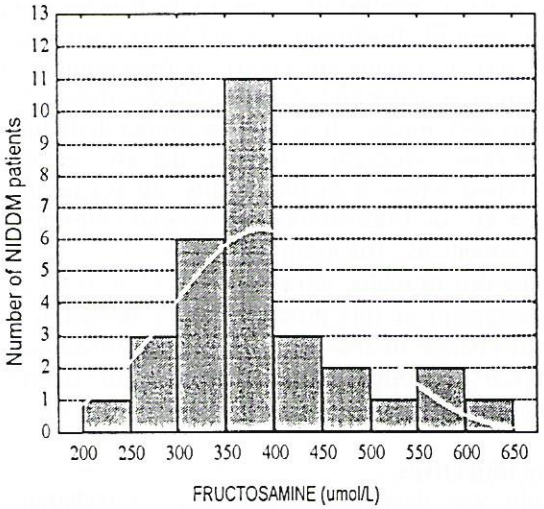

Fig3: Frequency distribution of NIDDM patients in fructosamine assay

Both these curves show that the maximum distribution of the patients is in the category of poor glycaemic control that is 11-13% for HbA_{1c} level and 350 - 450 µ mole/L in fructosamine assay.

Discussion

Our study demonstrates a correlation of high statistical significance between fructosamine and HbA_{1c}. Similar reports have been cited in previous studies by Hindle[10]. (r=0.5) and Islam[2] (r=0.41). Moreover, fructosamine assay is relatively inexpensive and reliable. Fructosamine assay will reduce the cost of the test by fifty percent

Fructosamine assay is far rapid and convenient to perform[8] and being simple this assay can be automated for mass screening[14]. It was found that fructosamine assay is most economical glycaemic parameter to be performed at large scale[3].

The only precaution which has to be taken in fructosamine assay is that the concentration of total serum proteins must be within normal limits[15]. Especially serum albumin should not be <30g/L[9]. In contrary to this HbA_{1c} is reported to be falsely high in iron deficiency anaemia, uraemia and increased fetal Hb levels; falsely low values in haemolytic disease, haemo-globinopathies (S,C,D) and pregnancy[16,17] HbA_{1c} estimation is contraindicated in these clinical conditions. In such cases Fructosamine assay is the method of choice.

Conclusion

Our study demonstrates that fructosamine levels correlate significantly with those of HbA_{1c} in glycaemic assessment of NIDDM patients. Fructosamine assay is simple, convenient, cost effective and reliable.

So we suggest a shift from HbA_{1c} to fructosamine assay in routine assessment for glycaemic control provided the serum albumin level is 30gm/L or more.

Moreover, such studies are required to be carried out to unmask the statistical data at different aspects of diabetic glycaemia and its estimation by fructosamine assay to make the laboratories more conscious and patients more familiar with this method.

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