Original Article

On The Analysis of Blood Glucose Levels of Diabetic Patients

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Abstract

Paired observation tests (Paired t-test, Wilcoxon signed ranked test and Sign test) were employed to determine if there is any significant difference between fasting blood sugar and oral glucose tolerance tests of 47 diabetic patients. First, Jarque-Bera (JB) test was done to check for normality and it showed that not all the mean differences were normally distributed. Two sample t-test was used to check whether the level of change in blood glucose for fasting patients and patients who had glucose intake are the same. The fasting blood sugar test and oral glucose tolerance test were done hourly on two different occasions. The results revealed no difference between blood glucose testing on arrival and blood glucose testing after resting for one hour. The level of change in blood glucose after two hours for fasting patients was not the same with the level of change two hours after glucose administration. It was concluded that a single fasting blood glucose test is sufficient to diagnose diabetes on a visit to the laboratory while oral glucose tolerance test conducted hourly is necessary to monitor the blood to know when the glucose level is going high or low.

Keywords: Diabetic patients; Blood glucose level; Paired Observation tests; Normality test; Diabetes

1. Introduction

Diabetes is a disease associated with high level of glucose (sugar) in the blood of the sufferer. It is very harmful to the body. Blood glucose testing involves measuring the levels of blood glucose after fasting and after glucose intake. Fasting blood sugar test is required to diagnose diabetes and Oral glucose tolerance test (OGTT), done after glucose intake, helps to monitor the glucose level in the blood of a patient. A hormone called insulin manages or regulates the glucose level in the body. When the body cannot produce enough insulin or when the insulin produced cannot work effectively owing to some other conditions, the blood sugar level rises causing diabetes. If it is too high or too low, medical attention will be needed.

Statistical tests such as Paired Observation tests (including parametric and Non-parametric tests) are useful for ascertaining if there is any significant mean difference between the two measurements (before and after administration of glucose). Also, two sample t-test is useful for checking whether the level of change in fasting blood sugar after two hours is the same with the level of change after administration of glucose. In paired observation, we have Univariate case and Multivariate cases. In Univariate case, two samples obtained are correlated and must be dependent. The two samples are reduced to one by working the differences between the paired observations. Here, the assumptions are: X and Y have a bivariate normal distribution and, X and Y are correlated [1].

The approach used in controlling diabetes is the blood glucose testing. This test measures the concentration of glucose in the blood. Blood glucose testing consist of fasting blood glucose and glucose intake measurements. The fasting blood sugar is used to diagnose diabetes while glucose intake is conducted to measure the blood glucose level. Oral glucose tolerance test (OGTT) measures first and second stage insulin response to glucose. It compares the level of glucose in the blood before and after administering glucose. After testing the blood glucose level in the patient fasting, 75grams of glucose dissolved in water is given to the patient and another test conducted at one hour and two hours' interval. A glucometer (glucose meter) is used to measure blood glucose and the unit of measurement is mg/dl or mmol/l. Blood glucose level outside the normal range may be an indicator of a medical condition. If the blood sugar measured is greater than 140gm/dl two hours after, it means the person is pre-diabetic. If it is greater than 199mg/dl two hours after, it means the person has full blown diabetes. If the result is 139mg/dl, it is considered as normal. The purpose of testing blood glucose several times per day is to discover when the blood glucose level is going too low or too high. This will help the doctor to timely take steps to prevent the occurrence of low or high blood glucose levels. The test becomes a warning to those who fall into the category of pre-diabetic and full blown diabetes as it enables them to change their lifestyle and cut down the intake of foods that has the capacity to increase blood glucose level. It is also beneficial in the sense that it gives information on whether the body can tolerate glucose or not.

It is the practice in hospitals that when a patient arrives, the patient is kept two hours before administration of glucose thereafter, the patient is kept for another two hours to monitor glucose level

[2]. The problem here is to ascertain whether the resting period of two hours is adequate or not and secondly, is the level of change after two hours for fasting glucose and the level of change two hours after glucose administration the same? The purpose of this study is to determine whether there is any significant difference between fasting blood sugar test and oral glucose tolerance test by performing a statistical analysis on blood glucose measurements obtained from the medical laboratory department of a University Teaching Hospital in Nigeria.

The specific objectives are to:

1. Determine statistically the adequate period of rest before the administration of glucose.
2. Determine whether the glucose level increased after one hour, two hours after glucose administration.
3. Determine if the level of change after two hours, for fasting glucose is the same with the level of change two hours after glucose administration.

There are many paired observation test statistics available but this research is limited to three paired observation tests (parametric Paired t-test, Non-parametric Wilcoxon ranked sign test and Sign test) only. Sign test of the median, Wilcoxon signed rank test and paired t-test, were performed to ascertain whether the level of change after one hour and two hour intervals for fasting glucose and after glucose administration, respectively was the same. The two sample t–tests was performed to check whether the level of change in blood glucose for fasting blood sugar test was the same with the test done two hours after glucose intake.

2. Review of Literature

2.1 Review of works on glucose test

Glucose, the simplest form of sugar, is the body's major source of energy while, glucose test measures the amount of glucose in the blood. A hormone called Insulin regulates blood glucose level in the body. According to Martel (2012) [3], Diabetes occurs when the body does not produce enough insulin thereby causing the blood glucose to rise, or when the body cannot use the insulin produced effectively. If diabetes is left untreated, the high blood glucose level will cause damages in the organs. People with diabetes normally go for blood glucose test to determine whether their condition is being managed well or not. A high blood glucose level is an indication of poor management of the diabetes. Blood glucose testing provides information that helps the physician to make decisions on diet and medication dosage.

Saudek *et al* (2006) [4], states that since there is increasing rate of diabetes, there is need to learn how to successfully control blood glucose level as this, leads to significant reduction in mortality due to diabetes, where assessing glycaemia in diabetes is challenging.

According to Haller *et al.* (2004) [5], blood glucose testing helps in administration of medication e.g., deciding the dosage to take. It also tells when the blood glucose level is high or low. An important benefit of blood sugar testing is that diabetic patients are sensitized to read and interpret their results thus, enabling them to take proper actions to improve control of the disease. People on insulin therapy or other medications should test their blood glucose level regularly so as to know when their blood glucose level is going too low or too high. Fasting blood sugar test is used to diagnose diabetes while Oral glucose tolerance test is conducted to measure blood glucose level. People with diabetes always have their blood sugar level higher than those without diabetes. According to American Diabetes Association (2006) [6], the normal level of blood glucose in diagnosis ranges from 126mg/dl to 140mg/dl.

2.2 Review of works on Paired Observation Tests

Given two random samples from two populations, we call the data paired if the j^{th} measurement of the first sample is paired with the k^{th} measurement of the second sample. Paired data or observations can be analyzed using parametric and non-parametric methods.

Conover (1999) [7], states that parametric test involves population distribution that has a particular form and also involves hypothesis about the population parameters, while non-parametric test does not make assumptions about the underlying distribution. Non-parametric tests use approximate solutions to a particular problem, while parametric tests use particular solutions to approximate problems. Hollander and Wolfe (1973) [8], posits that parametric test depends on distributional assumptions while non – parametric test do not require any strict distributional assumptions. They analyze ranks of a variable instead of using the original values.

Paired t – test is a parametric statistical method used to compare two population means that emanate from two samples that are correlated. According to Zaiontz (2013) [9], paired t – test is used to test for the differences in mean of two samples which are dependent on one another. In this case, the null hypothesis is such that the mean difference is equal to zero. Paired t – test involves comparing two means for information where every person who participated in one sample also participated in the other sample. The difference scores are created for every individual, and we compare mean differences between the results with the mean difference for the population [10,11].

Non-parametric tests for analyzing paired observations include the Sign Test and the Wilcoxon Signed Rank Test. Given n observations from a population with two samples u and v. For each observation, two paired samples $\{u_1...u_n\}$ and $\{v_1 . . . v_n\}$ will result. In conducting Wilcoxon signed rank test for paired samples where $Z_i = v_i – u_i$ for all $i = 1 . . . n$, it is required that Z_i are independent. For v_i and u_i, ranking can then be applied and differences taken. Also, the distribution of Z_i's is symmetric. The hypothesis, H_0 can be stated as "the distribution of difference scores in the population is symmetric about zero" [9]. The Wilcoxon signed rank test is used to test if the median of a symmetric population is zero. Firstly, the data is ranked without regarding the sign. Secondly, we attach the signs of the main observation to their corresponding ranks [12]. The test can also be applied in the comparison of two dependent or correlated data in which the measurements are ordinal and for samples that are matched or measurements that are repeated on a single sample to check whether the rank in their population mean differ or not [13,14]. In this case, the null hypothesis is that there is no difference in the two paired measurements. According to Mcdonald (2014) [15], the difference scores between the two paired values are computed then, the absolute differences from the smallest to the largest are taken from the measurements. After that, ranks are assigned to the values. Signs (+ or –) are also assigned according to the differences. The sum of positive and the negative ranks respectively, are then computed. The rank of all differences in one direction should be added and the smaller of those sums becomes the test statistic. The z distribution approximation for large samples is used and the statistical decision is to reject the null hypothesis if z calculated is greater than 1.96 at 0.05 level of significance.

The Sign test is a non-parametric method used to test the differences between pairs of observations. It is used to determine whether one of a pair of observation is greater or less than the other. It can also be used to test for trend in series of ordinal measurements or a test for correlation [16]. Sign test is used to test a claim that involves matched pairs of sample data, a claim involving numerical data with two categories, or a claim about the population median against a hypothesized value. In other to use it, the data values are first converted to 'plus' and 'minus' signs before testing. The sign test makes very few assumptions about the type of distributions under test. It tests the hypothesis that the differences can either be positive or negative using paired data. The test statistic for large samples is $(\text{plus} - \text{minus})^2 / (\text{plus} + \text{minus})$. 'Plus' is the number of positive values and 'minus' is the number of negative values. The hypothesis is "the positive and negative values are equally likely" and the test statistic follows chi square distribution with one degree of freedom [17]. Sign test is used to compare the differences between paired groups, analyzing only the signs of the difference scores. The null hypothesis is "the median difference is zero". If the null hypothesis is true, then approximately half of the differences will be positive and the other half will be negative. The test statistic is the smaller of the number of positive signs or negative signs. The critical value is determined using the Sign Test Table. The decision is to reject H_0 if the test statistic (i.e., the smaller of the positive or negative signs) is less than or equal to the critical value. The p-value is computed based on the observed test statistic.

The sign test is different from the Wilcoxon Signed rank test as the latter is rank based and considers the magnitude of the differences between the paired samples rather than their signs. Both tests differ from parametric t test and assumes that the distribution of the differences within pairs are symmetric without needing normality [8]. In paired samples, we are interested in testing whether the population median is equal or not. That is,

$$\begin{aligned} &H_0 : M_1 = M_2 \\ &\textit{against} \qquad \ldots\ldots\ldots\ldots\ldots\ldots(2.1) \\ &H_1 : M_1 \neq M_2 \end{aligned}$$

If the parametric test assumptions are satisfied, then paired sample t – test can be used to test the hypothesis.

Sign test can also be used to check if a binomial distribution has equal chance of success and failure. In this case, the differences of the paired values, using the signs of the individual differences, are taken. Then, the binomial distribution table is used for the test assuming probability, $p = 0.5$ when the sign is positive and $q = 0.5$ when the sign is negative [16]. Here, the null hypothesis is "the number of positive differences is approximately equal to the number of negative differences". Lucky and Bright (2012) [18], noted that Sign and Wilcoxon signed rank tests are most suitable for analyzing ranked or ordinal data which often deviate drastically from normality thus, making the use of parametric test unsuitable.

The sign test and Wilcoxon signed rank test have been used to analyze many real life occurrences. Ho, *et a*l. (2004) [19] used the Wilcoxon Signed-Rank Test to analyze the effect of a plastic implant into the soft palate of 12 chronic snorers to see if it would reduce the volume of snoring. The result showed that the median change in snoring volume was significantly different from zero. In another experiment, Buchwalder and Huber-Eicher (2004) [20] also used the Wilcoxon Signed-Rank Test to study the aggressive reactions of turkeys housed in pens towards unfamiliar turkey brought into the pen. They found that the median difference between the number of pecks per test in the small pen and in the large pen was significantly greater than zero.

3. Methods

Statistical analysis on blood glucose measurements of 47 Diabetic Patients obtained from the medical laboratory department of Nnamdi Azikiwe University Teaching Hospital, Nnewi, Anambra State, Nigeria was done using paired observation tests, and two sample t test. The Jarque-Bera (JB) test was used to test for normality of the sample data before the analysis. Definitions and methods of data analysis used are discussed in the following sub-sections. Minitab 18 statistical software was used for all the computations.

3.1 Jarque – Bera (JB) test

The JB test is based on the classical measures of skewness and kurtosis. It is a "goodness – of – fit" test used to check whether the sample data has the skewness and kurtosis matching the normal distribution. The JB statistic has a chi squared distribution with two degrees of freedom $\chi^2(2)$ [21].

The JB statistic [22] is given as:

$$JB = n\left(\frac{\gamma_1^2}{6} + \frac{\gamma_2^2}{24}\right) \approx \chi^2(2) \quad \text{..........(3.1)}$$

where,

γ_1 is the skewness, γ_2 is the kurtosis and n is the sample size.

$$\gamma_1 = \frac{\sum_{i=1}^{n}\left(d_i - \bar{d}\right)^3}{ns^3} \quad \text{.................(3.2)}$$

$$\gamma_2 = \frac{\sum_{j=1}^{n}\left(d_j - \bar{d}\right)^4}{ns^4} \quad \text{................(3.3)}$$

where,

d is the difference in each observation**,**

s is the standard deviation.

Equation (3.1) was used to test whether the differences obtain are normally distributed or not.

Hypothesis:

H_0: Data follows a standard normal distribution.

H_1: Data does not follow a standard normal distribution.

Decision rule: we reject $H_{0,}$ if JB > $\chi^2(2)$, otherwise accept H_0

3.2 Paired Observations Tests

In this section, we describe parametric and non-parametric methods used in the study. The methods are Paired t test, Wilcoxon Signed Rank Test and Sign Test for paired samples.

1. Paired t – test: Paired t – test matches responses which are dependent or which are related in a pairwise manner. The matching helps to detect variability that is between the pairs which usually result in small error terms thus, increasing the sensitivity of the hypothesis test or confidence interval. It is used to determine if there are any significant differences that exist between the mean values, under two different conditions but with the same measurement.

Therefore, the paired t-test hypothesis is:

H_0: There is no significant mean difference between the two paired samples.

H_1: There is significant mean difference between the two paired samples.

That is,

H_0: $\mu_d = 0$ ……………(3.4)

H_1: $\mu_d \neq 0$…………...(3.5)

where μ_d is estimated by the mean differences $\bar{d}$ between the fasting blood glucose test (X_i) and oral glucose tolerance test (Y_i); using α = 0.05

The t-statistic is given as:

$$t_{cal1} = \frac{\bar{d}}{s_d / \sqrt{n}} \quad \text{…………(3.6)}$$

where,

$$\bar{d} = \frac{\sum_{i=1}^{n} d_i}{n} \quad \text{………………..(3.7)}$$

And

$$S_d = \sqrt{\frac{\sum_i \left(d_i - \bar{d}\right)^2}{n-1}} \quad \text{…………(3.8)}$$

Using the confidence interval (CI),

$$CI = \bar{d} \pm \left(t_{\left(1-\frac{\alpha}{2}\right)} \times \frac{s_d}{\sqrt{n}} \right) \text{……………(3.9)}$$

Accept H_0, if the $100(1-\alpha)\%$ confidence internal (CI) for the mean difference include zero [17,23].

Note that,

$\bar{d}$ = Mean difference between paired samples.

μ_d = hypothesized population mean difference (usually 0).

S_d = standard deviation of paired differences.

d_i = sample item difference.

n = sample size (number of pairs in the sample).

n – 1 = degree of freedom for the test statistics.

t = paired sample t–test.

In other words, if t calculated is less than the t tabulated, we reject H_0 otherwise we accept the null hypothesis.

2. Assumptions of paired t-test

The test can only be performed with the matched pairs of sample. The differences of the paired sample must be normally distributed and the data must be continuous.

1. Wilcoxon Signed Ranked Test

Wilcoxon signed ranked test is useful in cases involving symmetric continuous distribution. In using it, we assume that the mean is equal to the median.

The null hypothesis is

$H_0: \mu = \mu_0$……………………(3.10)

Given the random samples $x_1, x_2, \ldots, x_n$ which come from a continuous and symmetric distribution with mean (median) = μ

1) Comparing the differences $|x_j - \mu_0|$ where j = 1, 2, . . . , n
2) We are to rank the absolute differences $|x_j - \mu_0|$ for j = 1, 2 , . . , n in ascending order
3) Signs + or – are allocated based on the direction of the differences, by computing the sum of the positive and the negative ranks respectively.
4) Denoting the sum of positive ranks by r^+ and the sum of negative ranks as r^-.

The test statistic is given as

$R = \min(r^+, r^-)$ ------------------(3.11)

where R is the Wilcoxon signed rank

If R calculated is less than or equal to tabulated R, we reject H_0 at 0.05 level of significance [17].

Assumptions of Wilcoxon Signed Rank test

The difference between the two data values of a pair is continuous and symmetric. The two samples need to be dependent observations of the cases and the data are measured on an ordinal or interval scale.

2. Sign test

Sign test, as had been discussed in section 2.1, is used to compare outcomes between paired groups, where only the signs of the difference scores are analyzed. The Probability value (p-value) is computed based on the observed test statistic; the test statistic follows a binomial distribution.

We have

$p(x \text{ is success}) = [n!/x!(n - X)!]\, p^x (1 - p)^{n-x}$…….(3.12)

where,

n = no of observation,

p is the probability of success or failure = 0.5.

x denotes the Sign test statistic [17].

Assumptions of Sign Test

The observation can be independent. It can be identically distributed and the measurement scale is at least ordinal.

Our interest was to determine whether the binomial distribution has equal chance of success and failure hence, we compared the outcome between fasting blood glucose test (X_i) and oral glucose tolerance test (Y_i); using α at 99%, 95% and 90% confidence level respectively.

3.3 Two sample t- test

Two sample t-test is a statistical method used to evaluate whether the means of two independent populations differ or not, the observation from one sample are not related to the observation from the other sample. The two sample t-test uses the sample standard deviations to estimate the variance, σ^2 for each population [24-26]. Reject the null hypothesis if the p-value is less than 0.05.

We compare the level of change in $d_3 = x_3 - x_1$ which has to do with two hours before glucose intake and the level of change in $d_4 = y_1 - x_3$ after glucose administration whether they are the same or not.

The hypothesis is

$$H_0 : \mu_{d_3} = \mu_{d_4} \text{ ....(3.13)}$$

$$H_1 : \mu_{d_3} \neq \mu_{d_4} \text{ ....(3.14)}$$

where,

d_3 = change in blood sugar after two hours for fasting patients.

d_4 = change in blood sugar after administration of glucose.

The point estimate for $\mu_{d_3} - \mu_{d_4}$ is $\bar{x}_{d_3} - \bar{x}_{d_4}$

where,

$$\bar{x}_{d_k} = \frac{\sum_{i=1}^{n} x_{d_k}}{n} \text{ ....(3.15)}$$

$$\bar{x}_{d_l} = \frac{\sum_{j=1}^{n} x_{d_l}}{n} \text{ ....(3.16)}$$

The standard deviation is

$$s_d = \sqrt{\frac{\sigma_1^{\ 2}}{n_1} + \frac{\sigma_2^{\ 2}}{n_2}} \text{ ....(3.17)}$$

The pooled variance becomes

$$s_p^2 = \frac{(n_1 - 1)s_1^2 + (n_2 - 1)s_2^2}{n_1 + n_2 - 2} \text{ .....(3.18)}$$

Then the test statistic becomes

$$t* = \frac{\bar{x}_{d_3} - \bar{x}_{d_4}}{s_p \sqrt{\frac{1}{n_1} + \frac{1}{n_2}}} \text{ ....(3.19)}$$

(Nduka, 2015) [23]with degrees of freedom equal

$$d_f = n_1 + n_2 - 2 \text{ ....(3.20)}$$

where,

s_d = the standard deviation of the differences.

$s_p^{\ 2}$ = the pooled sample variance from population 1 and population 2.

$s_1^{\ 2}$ = variance of the sample difference one.

$s_2^{\ 2}$ = variance of the sample difference two.

n_1 = size of the sample difference one.

n_2 = size of the sample difference two.

k is the third difference (d_3)

l is the fourth difference (d_4)

Assumptions of two sample t- test

The data are continuous, they follow normal distribution and variances of the two populations are equal. The two samples are independent and both are simple random samples from their respective population.

3.4 Definition and measurement of variables

The fasting variables were taken on two different occasions after the initial value, X_1 at one-hour and two-hour interval each. The glucose intake variables were also taking on two occasions at one-hour interval each. Paired Observation tests were used to check whether it is possible to take the recording on three occasions. For the fasting variables we denote the variables with x_1, x_2, and x_3.

where,

X_1 = blood glucose test on arrival.

X_2 = blood glucose after resting for one hour.

X_3 =blood glucose test after two hours.

On the other hand, we denote the glucose intake variables with y_1, y_2,

where,

y_1 =blood glucose test taken one hour after administration of glucose.

y_2 =blood glucose test taken two hours after administration of glucose.

For the fasting variables, the differences are:

$d_1 = x_2 – x_1$ (change in blood sugar after one hour for fasting patients)

$d_2 = x_3 – x_2$ (change in blood sugar for fasting patents at an interval of one –two hours)

$d_3 = x_3 – x_1$ (change in blood sugar level after two hours for fasting patients)

For the glucose intake variables, the differences are:

$d_4 = y_1 – x_3$ (change in blood sugar level after one hour of administration of glucose)

$d_5 = y_2 – x_3$ (change in blood sugar level after two hours of glucose administration).

$d_6 = y_2 – y_1$ (change in blood sugar level at one-hour and two-hour interval after glucose administration).

4. Results

The aim of this work is to determine whether there is any significant difference between fasting blood sugar test and oral glucose tolerance test. The fasting blood sugar test was done hourly on two different occasions, while the oral glucose test was done hourly on two different occasions also. Paired observation tests were done to determine whether it is necessary to test diabetic patients fasting blood sugar hourly up to two different times or not and glucose intake hourly up to two different times, using paired t-test (parametric), Wilcoxon signed rank test and Sign test (Non-parametric). Finally, two sample t-test was used to test whether the level of change after two hours for fasting glucose is the same as the level of change one hour after glucose intake. The results are given in the following subsections.

4.1 Jarque-Bera (JB) test Result

This test was used to test for normality of the data set differences stated in Section 3.1, for the fasting and glucose intake variables. The results are shown in Table 1.

Table 1: Jarque-Bera (JB) test Results

Variable	Skewness	Kurtosis	JB
d1	-0.11	2.41	11.46898
d2	0.5	1.42	5.907117
d3	0.51	-0.05	2.042346
d4	0.76	0.07	4.534129
d5	0.75	-0.05	4.411146
d6	2.82	10.04	259.6969

From the Table 1, $d_1 = 11.47$ and $d_6 = 259.7$ are both greater than $\chi^2_{(2)0.05} = 5.99$ while d_2, d_3, d_4 and d_5 are less than $\chi^2_{(2)0.05} = 5.99$. This implies that not all the data set differences are normally distributed (d_1 and

d_6 are not normally distributed) thus, indicating the use of both parametric and non-parametric tests.

4.2 Paired Observations Tests Results

Since the fasting variables are taking on two different occasions, we tested whether it is necessary to take the measurement at one-hour interval before administering glucose to the patient. Tables 2 to 4 summarizes the data set differences test results using the Paired t– test, Wilcoxon signed rank test and Sign test at 99%, 95% and 90% confidence intervals, respectively.

Table 2: Blood glucose test results at 99% Confidence Interval

99%	Paired t – test T statistic (p – value)	Wilcoxon signed rank test T statistic (p – value)	Sign test T statistic (p – value)
$d_1=x_2-x_1$	1.83 **(0.074)**	2.500 **(0.060)**	2.000 (0.0226)
$d_2=x_3-x_2$	4.18 (0.000)	5.000 (0.000)	5.000 (0.0000)
$d_3=x_3-x_1$	3.81 (0.000)	6.500 (0.001)	5.000 (0.0025)
$d_4=Y_1-x_3$	17.89 (0.000)	77.50 (0.000)	72.00 (0.0000)
$d_5=Y_2-x_3$	17.30 (0.000)	83.00 (0.000)	76.00 (0.0000)
$d_6=Y_2$-y_1	3.87 (0.000)	4.00 (0.000)	4.00 (0.0000)

Remark: If $p > 0.05$, we accept H_0

Table 3: Blood glucose test results at 95% Confidence Interval

95%	Paired t – test statistic (p – value)	Wilcoxon signed rank test statistic (p – value)	Sign test statistic (p–value)
$d_1=x_2-x_1$	1.83 (0.074)	2.500 (0.060)	2.000 (0.0226)
$d_2=x_3-x_2$	4.18 (0.000)	5.000 (0.000)	5.000 (0.000)
$d_3=x_3-x_1$	3.81 (0.000)	6.500 (0.001)	5.000 (0.0025)
$d_4=Y_1-x_3$	17.89 (0.000)	77.50 (0.000)	72.00 (0.0000)
$d_5=Y_2-x_3$	17.30 (0.000)	83.00 (0.000)	76.00 (0.0000)
$d_6=Y_2$-y_1	3.87 (0.000)	4.00 (0.000)	4.00 (0.0000)

Remark: If $p > 0.05$, we accept H_0

Table 4: Blood glucose test results at 90% Confidence Interval

90%	Paired t – test statistic (p – value)	Wilcoxon signed rank test statistic (p – value)	Sign test statistic (p – value)
$d_1=x_2-x_1$	1.83 (0.074)	2.500 (0.060)	2.000 (0.0226)
$d_2=x_3-x_2$	4.18 (0.000)	5.000 (0.000)	5.000 (0.0000)
$d_3=x_3-x_1$	3.81 (0.000)	6.500 (0.001)	5.000 (0.0025)
$d_4=Y_1-x_3$	17.89 (0.000)	77.50 (0.000)	72.00 (0.0000)
$d_5=Y_2-x_3$	17.30 (0.000)	83.00 (0.000)	76.00 (0.0000)
$d_6=Y_2-y_1$	3.87 (0.000)	4.00 (0.000)	4.00 (0.0000)

Remark: If $p > 0.05$, we accept H_0

The results (Tables 2 to 4), show that:

For paired t – test of fasting variables x_1 and x_2, the confidence interval for the mean difference between the two fasting variables include zero suggesting that there is no significant difference between them. The p–value is 0.074 further suggesting that the data are consistent with H_0: $\mu_d = 0$. Since $p > 0.05$, we accept H_0 and conclude that there is no difference between the two paired samples.

For Wilcoxon signed rank test, the fasting variables, x_1 and x_2 measurement results shows that p – value (p = 0.060) is greater than 0.05 and the median difference between x_1 and x_2 is zero. Therefore, we accept H_o and conclude that there is no significant difference in median of the fasting variables after one-hour interval. However, the sign test for the fasting variables, x_1 and x_2 shows that the p – value (p = 0.0226) is less than 0.05 implying difference between them hence, we reject H_0 and conclude that the fasting variables, x_1 and x_2 do not have common median.

4.3 Two sample t- test Result

From Section 3.4, the level of change after two hours for fasting glucose, $d_3 = x_3 - x_1$ and the level of change one hour after glucose administration, $d_4 = y_1 - x_3$. We computed the means and variances of both d_3 and d_4, and their pooled variance as discussed in Section 3.3. The results show that $\bar{x}_{d_3} = 7.5$, $\bar{x}_{d_4} = 73.8$, $s_{d_3} = 13.4$, $s_{d_4} = 30.5$, $s_p^2 = 23.5325$, p = 0.000 and $df = 92$. The p-value $_{cal}$ (0.000) is less than p-value $_{tab}$ (0.05), we reject H_0 and conclude that there is significant difference between the level of change in blood sugar for fasting patients and the level of change in blood sugar after administration of glucose.

5. Discussion

These results suggest that it is not necessary to take the blood glucose measurements every one hour rather, it should be taken every two hours since there was no difference between blood glucose tested on arrival and one hour after resting, as shown by the results of paired t – test and Wilcoxon signed rank test at all three levels of significance (99%, 95% and

90%) respectively. This implies that conducting fasting blood glucose test three different times on a visit, is not necessary as it amounts to a waste of time, money and resources. Furthermore, d_2 to d_6 in Tables 2 to 4, have p-values ($p = 0.00$) less than 0.05 for all the three paired-observation tests, indicating that there is significant difference between the variables. The change in blood sugar level after glucose intake is significant. The blood sugar level increased progressively after administering glucose at one-hour and two-hour intervals. This shows that a measurement in one hour or two after administering glucose is sufficient to give the diagnosis. There was significant difference between the change in blood sugar level after two hours for fasting glucose test and after one hour of glucose administration for the Oral glucose tolerance test. As a result, it is necessary to measure glucose level at hourly intervals after administering glucose to help monitor when the blood glucose level is too high or too low.

6. Conclusion

Based on the above results, it was concluded that a single fasting blood glucose test is sufficient to diagnose diabetes on a visit to the laboratory while oral glucose tolerance test conducted hourly is necessary to monitor the blood to know when the glucose level is going high or low.

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Appendix A

The Data (Blood Glucose Measurements) Used For The Statistical Analysis.

Fasting blood sugar variable Glucose intake variables

N/s	Initial value	1st hr	2nd hr	1st hr after taken glucose	2nd hr after taken glucose	3rd hr after taken glucose
1	90	93	98	140	145	152
2	85	80	87	152	152	160
3	100	95	102	181	190	195
4	110	105	100	200	225	210
5	109	100	103	171	180	182
6	92	105	130	185	183	180
7	108	110	110	165	168	170
8	88	100	120	161	165	170
9	115	118	124	181	180	181
10	116	115	100	200	205	205
11	93	100	97	215	217	220
12	84	92	96	150	145	140
13	82	86	90	162	160	160
14	91	92	94	135	140	142
15	100	102	100	140	140	150
16	112	100	107	170	180	182
17	120	125	128	200	250	320
18	100	126	130	195	200	202
19	128	120	125	195	190	198
20	130	124	110	205	207	209
21	101	74	85	200	210	212
22	92	85	88	140	145	160
23	78	75	80	140	152	155
24	90	90	98	142	150	152
25	87	93	97	170	171	171
26	90	103	108	185	184	180
27	85	95	90	190	192	200
28	119	120	119	198	200	210
29	102	95	100	200	210	220
30	96	98	104	250	280	282
31	92	94	100	200	225	210
32	83	92	100	200	205	205
33	99	101	98	215	217	220
34	107	109	101	162	160	160
35	83	94	103	135	140	145
36	105	110	125	175	180	185
37	118	115	120	195	200	205

38	100	100	105	185	188	190
39	80	85	102	240	242	250
40	77	81	110	245	252	260
41	88	100	120	180	182	188
42	82	85	90	195	195	199
43	100	125	130	216	220	222
44	120	125	128	184	189	200
45	116	116	120	235	240	241
46	110	105	108	270	272	275
47	92	105	130	202	210	210

Data Source: Nnamdi Azikiwe University Teaching Hospital Nnewi (NAUTH), 2019.

Appendix B

The differences data set for the fasting and glucose intake variables Minitab 18 result output of Skewness and Kurtosis (See JB's computation in Table 4.1).

Descriptive Statistics: d1, d2, d3, d4, d5, d6

Variable	Skewness	Kurtosis
d1	-0.11	2.41
d2	0.50	1.42
d3	0.51	-0.05
d4	0.76	0.07
d5	0.75	-0.05
d6	2.82	10.04

Appendix C

Paired Observation tests of Diabetic Patients ($\alpha = 99\%$)

Paired T-Test and CI: 1st hr, Initial value

Paired T for 1st hr - Initial value

	N	Mean	StDev	SE Mean
1st hr	47	101.234	13.795 2.012	
Initial value	47	98.830	13.921	2.031
Difference	47	2.40426	9.00642	1.31372

99% CI for mean difference: (-1.12573, 5.93424)

T-Test of mean difference = 0 (vs not = 0): T-Value = 1.83 P-Value = 0.074

Paired T-Test and CI: 2nd hr, 1st hr

Paired T for 2nd hr - 1st hr

	N	Mean	StDev	SE Mean
2nd hr	47	106.596	13.812	2.015
1st hr	47	101.234	13.795	2.012
Difference	47	5.36170	8.78592	1.28156

99% CI for mean difference: (1.91814, 8.80527)

T-Test of mean difference = 0 (vs not = 0): T-Value = 4.18 P-Value = 0.000

Paired T-Test and CI: 2nd hr, Initial value

Paired T for 2nd hr - Initial value

	N	Mean	StDev	SE Mean
2nd hr	47	106.596	13.812	2.015
Initial value	47	98.830	13.921	2.031
Difference	47	7.76596	13.97080	2.03785

99% CI for mean difference: (2.29023, 13.24169)

T-Test of mean difference = 0 (vs not = 0): T-Value = 3.81 P-Value = 0.000

Paired T-Test and CI: 1st hr after taken glucose, 2nd hr

Paired T for 1st hr after taken glucose - 2nd hr

	N	Mean	StDev	SE Mean
1st hr after tak	47	186.213	31.700	4.624
2nd hr	47	106.596	13.812	2.015
Difference	47	79.6170	30.514	4.4510

99% CI for mean difference: (67.6570, 91.5770)

T-Test of mean difference = 0 (vs not = 0): T-Value = 17.89 P-Value = 0.000

Paired T-Test and CI: 2nd hr after taken glucose, 2nd hr

Paired T for 2nd hr after taken glucose - 2nd hr

	N	Mean	StDev	SE Mean
2nd hr after tak	47	191.766	35.105	5.121
2nd hr	47	106.596	13.812	2.015
Difference	47	85.1702	33.7441	4.9221

99% CI for mean difference: (71.9445, 98.3959)

T-Test of mean difference = 0 (vs not = 0): T-Value = 17.30 P-Value = 0.000

Paired T-Test and CI: 2nd hr after taken glucose, 1st hr after taken glucose

Paired T for 2nd hr after taken glucose - 1st hr after taken glucose

	N	Mean	StDev	SE Mean
2nd hr after tak	47	191.766	35.105	5.121
1st hr after tak	47	186.213	31.700	4.624
Difference	47	5.55319	9.83076	1.43396

99% CI for mean difference: (1.70011, 9.40627)

T-Test of mean difference = 0 (vs not = 0): T-Value = 3.87 P-Value = 0.000

Wilcoxon Signed Rank Test: d1

Test of median = 0.000000 versus median not = 0.000000

	N for Wilcoxon	Estimated	N Test Statistic	P	Median
d1	47	44	656.5	0.060	2.500

Wilcoxon Signed Rank Test: d2

Test of median = 0.000000 versus median not = 0.000000

	N for Wilcoxon	Estimated	N Test Statistic	P	Median
d2	47	46	910.0	0.000	5.000

Wilcoxon Signed Rank Test: d3

Test of median = 0.000000 versus median not = 0.000000

	N for Wilcoxon	Estimated	N Test Statistic	P	Median
d3	47	45	825.0	0.001	6.500

Wilcoxon Signed Rank Test: d4

Test of median = 0.000000 versus median not = 0.000000

	N for Wilcoxon	Estimated	N Test Statistic	P	Median
d4	47	47	1128.0	0.000	77.50

Wilcoxon Signed Rank Test: d5

Test of median = 0.000000 versus median not = 0.000000

	N for Wilcoxon	Estimated	N Test Statistic	P	Median
d5	47	47	1128.0	0.000	83.00

Wilcoxon Signed Rank Test: d6

Test of median = 0.000000 versus median not = 0.000000

	N for Wilcoxon	Estimated	N Test Statistic	P	Median
d6	47	44	870.0	0.000	4.000

Sign Test for Median: d1

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d1	47	14	3	30	0.0226	2.000

Sign Test for Median: d2

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d2	47	9	1	37	0.0000	5.000

Sign Test for Median: d3

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d3	47	12	2	33	0.0025	5.000

Sign Test for Median: d4

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d4	47	0	0	47	0.0000	72.00

Sign Test for Median: d5

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d5	47	0	0	47	0.0000	76.00

Sign Test for Median: d6

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d6	47	8	3	36	0.0000	4.000

Appendix D

Paired Observation tests of Diabetic Patients (α = 95%)

Paired T-Test and CI: 1st hr, Initial value

Paired T for 1st hr - Initial value

	N	Mean	StDev	SE Mean
1st hr	47	101.234	13.795	2.012
Initial value	47	98.830	13.921	2.031
Difference	47	2.40426	9.00642	1.31372

95% CI for mean difference: (-0.24013, 5.04864)

T-Test of mean difference = 0 (vs not = 0): T-Value = 1.83 P-Value = 0.074

Paired T-Test and CI: 2nd hr, 1st hr

Paired T for 2nd hr - 1st hr

	N	Mean	StDev	SE Mean
2nd hr	47	106.596	13.812	2.015
1st hr	47	101.234	13.795	2.012
Difference	47	5.36170	8.78592	1.28156

95% CI for mean difference: (2.78206, 7.94135)

T-Test of mean difference = 0 (vs not = 0): T-Value = 4.18 P-Value = 0.000

Paired T-Test and CI: 2nd hr, Initial value

Paired T for 2nd hr - Initial value

	N	Mean	StDev	SE Mean
2nd hr	47	106.596	13.812	2.015
Initial value	47	98.830	13.921	2.031
Difference	47	7.76596	13.97080	2.03785

95% CI for mean difference: (3.66398, 11.86794)

T-Test of mean difference = 0 (vs not = 0): T-Value = 3.81 P-Value = 0.000

Paired T-Test and CI: 1st hr after taken glucose, 2nd hr

Paired T for 1st hr after taken glucose - 2nd hr

	N	Mean	StDev	SE Mean
1st hr after tak	47	186.213	31.700	4.624
2nd hr	47	106.596	13.812	2.015

Difference	47	79.6170	30.5148	4.4510

95% CI for mean difference: (70.6575, 88.5765)

T-Test of mean difference = 0 (vs not = 0): T-Value = 17.89 P-Value = 0.000

Paired T-Test and CI: 2nd hr after taken glucose, 2nd hr

Paired T for 2nd hr after taken glucose - 2nd hr

	N	Mean	StDev	SE Mean
2nd hr after tak	47	191.766	35.105	5.121
2nd hr	47	106.596	13.812	2.015
Difference	47	85.1702	33.7441	4.9221

95% CI for mean difference: (75.2626, 95.0779)

T-Test of mean difference = 0 (vs not = 0): T-Value = 17.30 P-Value = 0.000

Paired T-Test and CI: 2nd hr after taken glucose, 1st hr after taken glucose

Paired T for 2nd hr after taken glucose - 1st hr after taken glucose

	N	Mean	StDev	SE Mean
2nd hr after tak	47	191.766	35.105	5.121
1st hr after tak	47	186.213	31.700	4.624
Difference	47	5.55319	9.83076	1.43396

95% CI for mean difference: (2.66677, 8.43961)

T-Test of mean difference = 0 (vs not = 0): T-Value = 3.87 P-Value = 0.000

Wilcoxon Signed Rank Test: d1

Test of median = 0.000000 versus median not = 0.000000

	N for Wilcoxon	Estimated	N Test Statistic	P Median
d1	47	44	656.5 0.060	2.500

Wilcoxon Signed Rank Test: d2

Test of median = 0.000000 versus median not = 0.000000

	N for Wilcoxon	Estimated	N Test Statistic	P Median
d2	47	46	910.0 0.000	5.000

Wilcoxon Signed Rank Test: d3

Test of median = 0.000000 versus median not = 0.000000

	N for	Wilcoxon	Estimated	N Test Statistic	P Median
d3	47	45	825.0 0.001	6.500	

Wilcoxon Signed Rank Test: d4

Test of median = 0.000000 versus median not = 0.000000

	N for	Wilcoxon	Estimated	N Test Statistic	P Median
d4	47	47	1128.0 0.000	77.50	

Wilcoxon Signed Rank Test: d5

Test of median = 0.000000 versus median not = 0.000000

	N for	Wilcoxon	Estimated	N Test Statistic	P Median
d5	47	47	1128.0 0.000	83.00	

Wilcoxon Signed Rank Test: d6

Test of median = 0.000000 versus median not = 0.000000

	N for	Wilcoxon	Estimated	N Test Statistic	P Median
d6	47	44	870.0 0.000	4.000	

Sign Test for Median: d1

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P Median
d1	47	14	3 30	0.0226	2.000

Sign Test for Median: d2

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal Above	P Median
d2	47	9 1	37 0.0000	5.000

Sign Test for Median: d3

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal Above	P Median
d3	47	12 2	33 0.0025	5.000

Sign Test for Median: d4

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal Above	P Median
d4	47	0 0	47 0.0000	72.00

Sign Test for Median: d5

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d5	47	0	0	47	0.0000	76.00

Sign Test for Median: d6

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d6	47	8	3	36	0.0000	4.000

Appendix E

Paired Observation tests of Diabetic Patients (α = 90%)

Paired T-Test and CI: 1st hr, Initial value

Paired T for 1st hr - Initial value

	N	Mean	StDev	SE Mean
1st hr	47	101.234	13.795	2.012
Initial value	47	98.830	13.921	2.031
Difference	47	2.40426	9.00642	1.31372

90% CI for mean difference: (0.19896, 4.60955)

T-Test of mean difference = 0 (vs not = 0): T-Value = 1.83 P-Value = 0.074

Paired T-Test and CI: 2nd hr, 1st hr

Paired T for 2nd hr - 1st hr

	N	Mean	StDev	SE Mean
2nd hr	47	106.596	13.812	2.015
1st hr	47	101.234	13.795	2.012
Difference	47	5.36170	8.78592	1.28156

90% CI for mean difference: (3.21040, 7.51300)

T-Test of mean difference = 0 (vs not = 0): T-Value = 4.18 P-Value = 0.000

Paired T-Test and CI: 2nd hr, Initial value

Paired T for 2nd hr - Initial value

	N	Mean	StDev	SE Mean
2nd hr	47	106.596	13.812	2.015

Initial value	47	98.830	13.921	2.031
Difference	47	7.76596	13.97080	2.03785

90% CI for mean difference: (4.34510, 11.18682)

T-Test of mean difference = 0 (vs not = 0): T-Value = 3.81 P-Value = 0.000

Paired T-Test and CI: 1st hr after taken glucose, 2nd hr

Paired T for 1st hr after taken glucose - 2nd hr

	N	Mean	StDev	SE Mean
1st hr after tak	47	186.213	31.700	4.624
2nd hr	47	106.596	13.812	2.015
Difference	47	79.6170	30.5148	4.4510

90% CI for mean difference: (72.1452, 87.0888)

T-Test of mean difference = 0 (vs not = 0): T-Value = 17.89 P-Value = 0.000

Paired T-Test and CI: 2nd hr after taken glucose, 2nd hr

Paired T for 2nd hr after taken glucose - 2nd hr

	N	Mean	StDev	SE Mean
2nd hr after tak	47	191.766	35.105	5.121
2nd hr	47	106.596	13.812	2.015
Difference	47	85.1702	33.7441	4.9221

90% CI for mean difference: (76.9077, 93.4327)

T-Test of mean difference = 0 (vs not = 0): T-Value = 17.30 P-Value = 0.000

Paired T-Test and CI: 2nd hr after taken glucose, 1st hr after taken glucose

Paired T for 2nd hr after taken glucose - 1st hr after taken glucose

	N	Mean	StDev	SE Mean
2nd hr after tak	47	191.766	35.105	5.121
1st hr after tak	47	186.213	31.700	4.624
Difference	47	5.55319	9.83076	1.43396

90% CI for mean difference: (3.14605, 7.96033)

T-Test of mean difference = 0 (vs not = 0): T-Value = 3.87 P-Value = 0.000

Wilcoxon Signed Rank Test: d1

Test of median = 0.000000 versus median not = 0.000000

N for Wilcoxon Estimated N Test Statistic P Median

d1 47 44 656.5 0.060 2.500

Wilcoxon Signed Rank Test: d2

Test of median = 0.000000 versus median not = 0.000000

N for Wilcoxon Estimated N Test Statistic P Median

d2 47 46 910.0 0.000 5.000

Wilcoxon Signed Rank Test: d3

Test of median = 0.000000 versus median not = 0.000000

N for Wilcoxon Estimated N Test Statistic P Median

d3 47 45 825.0 0.001 6.500

Wilcoxon Signed Rank Test: d4

Test of median = 0.000000 versus median not = 0.000000

N for Wilcoxon Estimated N Test Statistic P Median

d4 47 47 1128.0 0.000 77.50

Wilcoxon Signed Rank Test: d5

Test of median = 0.000000 versus median not = 0.000000

N for Wilcoxon Estimated N Test Statistic P Median

d5 47 47 1128.0 0.000 83.00

Wilcoxon Signed Rank Test: d6

Test of median = 0.000000 versus median not = 0.000000

N for Wilcoxon Estimated N Test Statistic P Median

d6 47 44 870.0 0.000 4.000

Sign Test for Median: d1

Sign test of median = 0.00000 versus not = 0.00000

N Below Equal Above P Median

d1 47 14 3 30 0.0226 2.000

Sign Test for Median: d2

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d2	47	9	1	37	0.0000	5.000

Sign Test for Median: d3

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d3	47	12	2	33	0.0025	5.000

Sign Test for Median: d4

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d4	47	0	0	47	0.0000	72.00

Sign Test for Median: d5

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d5	47	0	0	47	0.0000	76.00

Sign Test for Median: d6

Sign test of median = 0.00000 versus not = 0.00000

	N	Below	Equal	Above	P	Median
d6	47	8	3	36	0.0000	4.000

Appendix F

Two sample t- test between $d_3 = x_3 - x_1$ and $d_4 = y_1 - x_3$,

Mean of d3 =(x_3-x_1)

Mean of d3 = 7.46809

Standard Deviation of d3 =(x_3-x_1)

Standard deviation of d3 = 13.3853

Mean of d4 =(y_1-x_3)

Mean of d4 = 73.7872

Standard Deviation of d4 =(y_1-x_3)

Standard deviation of d4 = 30.4695

Two-Sample T-Test and CI: d3, d4

Two-sample T for d3 vs d4

	N	Mean	StDev	SE Mean
d3	47	7.5	13.4	2.0
d4	47	73.8	30.5	4.4

Difference = mu (d3) - mu (d4)

Estimate for difference: -66.3191

95% CI for difference: (-75.9604, -56.6779)

T-Test of difference = 0 (vs not =): T-Value = -13.66, P-Value = 0.000, DF = 92

Both use Pooled StDev = 23.5325