بسم الله الرحمن الرحيم

Sudan University of Science and Technology

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Evaluation of Glucose Level in Serum and Plasma Using Different Anticoagulants at Room Temperature

تقويم مستوي سكر الجلكوز في مصل وبلازما الدم باستخدام موانع تجلط مختلفة في درجة حرارة الغرفة

A dissertation Submitted in Partial Fulfillment for the Requirement of M.Sc. Degree in Medical Laboratory Sciences (Clinical Chemistry)

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الاية

قال تعالى :

اقْرَأْ بِاسْمِ رَبِّكَ الَّذِي خَلَقَ (1) خَلَقَ الْإِنْسَانَ مِنْ عَلَقٍ (2) اقْرَأْ وَرَبُّكَ الْأَكْرَمُ (3) الَّذِي عَلَّمَ بِالْقَلَمِ (4) عَلَّمَ الْإِنْسَانَ مَا لَمْ يَعْلَمْ (5)

صدق الله العظيم

سورة العلق الآيات من (1-5)

Dedication

I dedicate this dissertation with love

To my grate father the best teacher I ever had

To my dear mother I hope when you look at me much proud you always see

To my sweetie sister and my supportive brothers

To my friends, coworkers and my family which they are volunteer for the blood samples

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To Allah at first to give me health and patience to complete this study

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Abstract

Back ground: Glucose is key to keeping the mechanisms of the body in top working order. Along with fats, glucose is one of the body's preferred sources of fuel. Glucose levels that are unhealthy or out of control can have permanent and serious effects.

Objective: To measure blood glucose level storage in different anticoagulants at room temperature.

Methodology: The study was conducted in Khartoum state from the period of March to September 2020. Including 60 healthy volunteers age range between 20 to 60 years old. The blood samples were dispended into different containers (fluoride oxalate, sodium citrate, lithium heparin and plain container). The glucose level was measured initially (at zero time) after 2, 4, 6 and 8 hours in the plasma and serum using colorimetric method (glucose oxides) then the data obtained were analyzed using (SPSS) in computer program.

Results: The result show that the stability of blood glucose level in fluoride oxalate up to 4 hours (P-value $\geq$ 0.005) while in heparin and sodium citrate and serum up to 2 hours (P-value $\geq$ 0.005).

Conclusion: From the result it was conclude that fluoride oxalate is more suitable anticoagulant for blood glucose estimation.

المستخلص

الخلفيه: الجلكوز هو المفتاح الذي يحافظ علي اليات الجسم في افضل حالة عمل . يعتبر الجلكوز و الدهون احد المصادر المفضلة للوقود في الجسم.

يمكن ان يكون لمستويات الجلكوز غير الطبيعيه ذات اثار دائمه و معقده.

الاهداف: قياس مستوي الجلكوز في بلازما ومصل الدم باستخدام موانع تجلط مختلفه في درجه حرارة الغرفه .

الطريقه: اجريت هذه الدراسه في ولايه الخرطوم في الفترة ما بين شهر مارس الي شهر سبتمبر 2020 . وتضمنت 60 متطوع اعمارهم ما بين 20 الي 60 سنه. اخذت عينه دم من كل متطوع ووزعت علي انابيب اختبار مختلفه (اوكسلات الفلور , سترات الصوديوم ، الهبرين وانبوب غير محتوي لمانع تجلط). وتم قياس سكر الجلكوز فورا بعد اخذ العينه وبعد ساعتان و اربعه ساعات وسته ساعات و ثمانيه ساعات .

وتم قياس الجلكوز باستخدام الكلروميتر (اكسده الجلكوز) و البيانات المتحصل عليها تم تحليلها بواسطه برنامج محوسب (الحزمة الاحصائيه للعلوم الاجتماعيه) .

النتائج: اظهرت النتائج ان استقرار مستوي الجلكوز في اوكسلات الفلور الي اربعه ساعات (القيمه المعنويه $0.005 \geq$) في حين ان الهبرين و سترات الصوديوم و مصل الدم يحفظ لمده ساعتين (القيمه المعنويه $0.005 \geq$).

الخلاصه: من النتائج يظهر ان اوكسلات الفلور هي افضل مانع تجلط لقياس الجلكوز.

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Chapter One
Introduction

1. Introduction, Rationale and Objectives

1.1 Introduction:

Blood glucose the main sugar that the body makes from the food in the diet. Glucose is carried through the bloodstream to provide energy to all cells in the body. Cells cannot use glucose without the help of insulin. Glucose is a simple sugar a monosaccharide. The body produces it from protein, fat and, in largest part, carbohydrate. Ingested glucose is absorbed directly into the blood from the intestine and results in a rapid increase in blood glucose. Glucose is also known as dextrose (William., 2018).

Blood glucose concentration depends on the relative rate of influx of glucose into the circulation and of its utilization. Its concentration is normally subject to rigorous control, rarely falling below 2.5 mmol/L at any time or rising above 80 mmol/L in healthy subjects after a meal or above 5.2 mmol/L after an overnight fast. Following a meal, glucose is stored as glycogen, which is mobilized during fasting state (Marshal, *et al*., 2012).

The plasma concentration of glucose is controlled by a number of hormones, in particular, insulin and glucagon. The physiology of glucose homeostasis is controlled primary by insulin release in response to elevated glucose levels. Glucose that is not needed for energy is stored in the form of glycogen as a source of potential energy, readily available

When needed. Most glycogen is stored in the liver and in muscle cells. When these and other body cells are saturated with glycogen, excess glucose is converted to fat and is stored as adipose tissue (Jorg and Thomas., 2013).

Anticoagulants are an additive that prevents the blood from clotting, in laboratory used in blood collection tubes. Most tube contains. (Giri., 2020).

Anticoagulants are portion of blood called plasma which is used for estimation of various substances in the blood (Hoffbrand *et al*., 2006).

1.2 Rationale

Plasma glucose measurement is most important for the screening, diagnosis, and monitoring for diabetes and metabolic dysregulation presents in conditions such as metabolic syndrome. These conditions result in pathological hyperglycemia or high glucose levels. Millions of people in the world have diabetes; hence, adequate monitoring of blood glucose in high-risk or pre-diabetic individuals can help guide lifestyle interventions that effectively reduce the risk of becoming diabetic.

The purpose of the study to help in choose the right anticoagulant that most suitable for accurate glucose estimation in routine laboratory setting and stable for long period term.

In Sudan there are no published studies showing the effects of storage of blood at room temperature with different anticoagulants.

1.3 Objective

1.3.1 General Objective

To assess blood glucose level in different anticoagulant at room temperature.

1.3.2 Specific Objective

1- To estimate blood glucose in different anticoagulants.

2-To correlate between blood glucose level and duration of storage according to type of anticoagulant.

Chapter Two

Literature Review

Chapter Two

2. Literature Review

2.1 Carbohydrate

The carbohydrate are a group of naturally occurring carbonyl compounds (aldehydes or ketones) that also contain several hydroxyl groups it may also include their derivatives which produce such compounds on hydrolysis. They are the most abundant organic molecules in nature and also referred to as "saccharides". The carbohydrates which are soluble in water and sweet in taste are called as "sugars". Carbohydrates consist of carbon, hydrogen and oxygen, the building blocks of all carbohydrates are simple sugars called monosaccharides (Lehninger, *et al*., 2000).

2.1.1 Classification of carbohydrate

Carbohydrates are classified as:

A. Monosaccharide: the most basic fundamental unit of a carbohydrate. These are simple sugars with the general chemical structure of C6H12O6). Theses easily are utilized for energy, causing a rapid rise in blood suger and insulin secretion from the pancreas.

Examples: fructose, lactose, maltose, sucrose, glucose, galactose and ribose.

B. Disaccharides: Compound sugars contain two monosaccharide with the elimination of a water molecule with the general chemical structure (C12H22O11) .Example: sucrose, lactose.

C. oligosaccharides: the polymer contains three to ten monosaccharides Example: maltodextrins, raffinose.

D. polysaccharides: polymers containing long chain of monosaccharide connected through glcosidic bonds.

These take longer to digest and therefore have a more gradual effect on the increase in blood sugar.

Examples: cellobiose, cellulose, amylose, glycogen and dextrin.

Starches: complex carbohydrates contain a large number of glucose molecules. Plants produce these polysaccgarides.

Examples include potatoes, chickpeas, pasta and wheat.

Fiber: non-digestible complex carbohydrates that encourage healthy bacterial growth in the colon and act as a bulking agent, easing defecaton. The main components include cellulose, hemicelluloses and pactin (Holesh, *et al.,* 2020).

2.1.2 Carbohydrate Metabolism

Carbohydrate digestion begins in the mouth, where the enzyme salivary amylase begins to breakdown complex sugars into monosaccharides. These can then be transported across the intestinal membrane into the blood stream and then to body tissues. In the cells, glucose, a six carbon sugar, is processed through a sequence of reactions into smaller sugars, and the energy stored inside the molecule is released. The first step of carbohydrate catabolism is glycolysis, which produces pyruvate, NADH, and ATP. Under anaerobic conditions, the pyruvate can be converted into lactate to keep glycolysis working. Under aerobic conditions, pyruvate enters the Krebs cycle, also called the citric acid cycle or tricarboxylic acid cycle.

In addition to ATP, the Krebs cycle produces high energy FADH2 and NADH molecules, which provide electrons to the oxidative phosphorylation process that generates mor high energy ATP molecules. For each molecules of glucose that is processed in glycolysis, a net of 36 ATPs can be created by aerobic respiration (Lumen Learning).

2.2 Glucose

Glucose is monosaccharide sugar that our bodies obtained from food and used as our principle energy source. The basic molecular form of glucose is C6H12O6. Glucose enters our body in several different forms such as fructose and galactose, which are monosaccharide and isomers of glucose. This monosaccharide can combine to form disaccharide such as lactose and fructose. Larger polymers of glucose are the polysaccharide forms of glucose which include starch, glycogen, and cellulose (Gurug and Jialal., 2019).

Circulating glucose is derived from three sources: intestinal absorption during the fed state, glycogenolysis, and gluconeogenesis. The major determinant of how quickly glucose appears in the circulation during the fed state is the rate of gastric emptying. Other sources of circulating glucose are derived chiefly from hepatic processes (Wallum., 1992).

2.2.1 Glucose metabolism

The main biochemical reaction employing glucose as its substrate is glycolysis, which, used by all tissues for the breakdown of glucose, provides energy in the form of adenosine triphosphate (ATP) and produces intermediates for other metabolic pathways. Since virtually all sugars are ultimately convertible to glucose, glycolysis serves as the hup of carbohydrate metabolism. In cells with mitochondria and an adequate supply of oxygen, pyruvate emerges as the end product of glycolysis via a 10-reaction series known as aerobic glycolysis.

Pyruvate is subsequently converted through oxidative decarboxylation into acetyl coenzyme A, the major fuel for the citric acid cycle. Alternatively, glucose undergoes anaerobic glycolysis to form lactate (Harvey and Ferrier., 2011).

2.2.2 Glucose Regulation

Glucoregulatory hormones include insulin, glucagon, amylin, GLP-1, glucose-dependent insulinotropic peptide (GIP), epinephrine, cortisol, and growth hormone. Of theses, insulin and amylin are derived from the β-cells, glucagon from the α-cells of the pancreas, and GLP-1 and GIP from the L-cells of the intestine. The glcoregulatory hormones of the body designed to maintain circulating glucose concentrations in a relatively narrow range (Gerich., 1993).

2.2.3 Metabolic Disorder

2.2.3.1 Hyperglycemia

Hyperglycemia is blood glucose greater than 125 mg/dl while fasting is greater than 180 mg/dl. A patient has impaired glucose tolerance, or pre-diabetes, with fasting plasma glucose of 100 mg/dl to 125 mg/dl. A patient is termed diabetic with fasting blood glucose of greater than 125 mg/dl (Hammer, *et al*., 2018) (Villegas-Valverde, *et al*., 2018).

When hyperglycemia is left untreated, it can lead to many serious life-threatening complications that include damage to the eye, kidneys, nerves, heart and the peripheral vascular system. Thus, it is vital to mange hyperglycemia efficiently to prevent complications of the disease and improve patient outcomes (Yari, *et al*., 2020).

Factor contributing to hyperglycemia includes reduced insulin secretion, degrease glucose utilization, and increase glucose production. Glucose homeostasis is a balance between hepatic glucose production and peripheral glucose uptake and utilization.

Insulin is the most important regulator of glucose homeostasis (Simon and Wittmann., 2019).

Diabetes mellitus: is a group of metabolic diseases characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both. The chronic hyperglycemia of diabetes is associated with long-termed damage, dysfunction, and failure of different organs, especially the eyes, kidneys, nerves, heart, and blood vessels (American Diabetes Association., 2010).

The majority of cases of diabetes can be broadly classified into 2

Categories: type 1 diabetes encompasses diabetes that is primarily a result of pancreatic beta cell destruction with consequent insulin deficiency, which is prone to ketoacidosis. This form includes cases due to an autoimmune process and those for which the etiology of beta cell destruction is unknown. Type 2 diabetes, may range from predominant insulin resistance with relative insulin deficiency to a predominant secretory defect with insulin resistance. Ketosis is not as common. Although some cases are difficult to classify. Gestational diabetes refers to glucose intolerance with onset or first recognition during pregnancy. Other specific types include a wide variety of relatively uncommon conditions, primarily specific genetically define forms of diabetes or diabetes associated with other diabetes or drug use (Zubin, *et al*., 2018).

2.2.3.2 Hypoglycemia

Is often defined by a plasma glucose concentration below 70 mg/ml; however, signs and symptoms may not occur until plasma glucose concentration drop below 55 mg/ml. glucose is the primary metabolic fuel for the brine under physiologic condition unlike other tissue of the body, the brine is very limited in supplying its glucose.

Expectedly, the brain requires a steady supply of arterial glucose for adequate metabolic function. Potential complications can arise from an interruption in the glucose supply.

As such, protective mechanisms to guard against low serum blood glucose (hypoglycemia) have evolved in the body. During fasting state, serum glucose level is maintained via gluconeogenesis and glycogenolysis in the liver. Gluconeogenesis is the pathway in which the glucose is generated from non-carbohydrate source.

These noncarbohydrate sources could be protin, lipids, pyruvate or lactate. In contrast, glycogenolysis is the breakdown of glycogen into glucose product. Much of glycogenolysis occurs in hepatocytes (liver) and myocytes(muscle) (Philip, *et al*., 2020).

2.2.4 Method of glucose measurement

2.2.4.1 Meter use

Use of glucose meter is common in physician offices or by patients to monitor blood glucose levels and establish patterns of glucose. Fluctuations over time with regular use and recording. Meters require a drop of blood applied to a test strip that inserts into a meter to estimate the plasma glucose level. The simple use of the meter can also help screen for acute hypoglycemia or hyperglycemic episodes and help patients plan meals, activities and insulin treatment (Pickering and Marsden., 2014).

2.2.4.2 Laboratory assessment

Current laboratory recommendations for plasma glucose measurements are to draw fasting blood samples in the morning rather than later in the day, as glucose levels tend to be higher in the morning than the afternoon. This sample should be placed on ice to minimized glycolysis and quicly processed (via plasma separation within 60 minutes) if plasma separation cannot occur within 60 minutes, the lab tech can add glycolytic inhibitor such as fluoride (Kim., 2016).

Fasting blood glucose should be obtained in the morning after an approximately 8 – to 10- hour fast (not longer than 16 h). Cerebrospinal fluid and urine can also be analyzed (Bishop, *et al*., *2018*).

Oral glucose tolerance test requires fasting blood glucose measurement in the morning. After the measurement, the patient receives oral glucose (usually a glucose load of 75g anhydrous glucose dissolved in water) that the patient consumes. The plasma glucose levels are measured again at 1-hour and 2-hours to analyze the glucose level changes (Sacks, *et al*., 2011).

Glycated hemoglobin HbA1c analysis provides evidence about an individual's average blood glucose level during the previous two to three months, which is predicated half-life of red blood cells (Khan and Weinstock., 2011).

Glycated hemoglobin should do every 3 months to monitor glycemic control and adjust the therapy (Grundy, *et al.*, 1999).

2.3 Anticoagulants

An agent that is use to prevent the formation of blood clots. Anticoagulants have various uses. Some are use for the prevention or treatment of disorder characterized by abnormal blood clots and emboli (William., 2018).

2.3.1 Purpose of Anticoagulants

Anticoagulants in laboratory used in blood collection tubes. Most tube contains an additive that prevents the blood from clotting. (Giri., 2020).

Blood is a combination of formed elements (RBCs, WBCs, and Platelets) in a liquid portion called plasma. These are used to prepare the whole blood or plasma during the collection of blood samples.

There is a difference in the plasma and in the serum for estimation of various substances in the blood (Hoffbrand., *et al.* 2006).

2.3.2 Examples of anticoagulants

2.3.2.1 Fluoride oxalate

It is Potassium oxalate and Sodium fluoride. Sodium fluoride acts as an antiglycolytic agent to ensure that no further glucose breakdown occurs within the sample after it is taken. Potassium oxalate removes calcium and acts as an anticoagulant. Used in clinical chemistry testing especially glucose (sugar) and lactate, glucose tolerance test (Giri.,2020).

2.3.2.2 Heparin

This is theoretically the best anticoagulant because it is a normal component of the blood and does not introduced any foreign contaminants to the blood specimen. Heparin accelerates the action of antithrombin III which neutralizes thrombin thus prevent the formation of fibrin from fibrinogen. It forms the complex of thrombin, antithrombin cofactor, and heparin, and prevents fibrin clot formation. It prevents the coagulation for 24 hours by neutralizing the thrombin, thus preventing the formation of fibrin clot from the fibrinogen. Heparin is added 0.2mg/ml of blood in each tube. This is best anticoagulant used for the estimation of PH, blood gases, electrolytes and ionized calcium (Michael., 2008).

2.3.2.3 Sodium Citrate

Citrate is used as trisodium citrate salt. Sodium citrate is widely used for coagulation studies for PT and PTT. The sample can be used for ESR by the westergren method. It will prevent the rapid deterioration of labile coagulation factors like factor V and factor VII.

(Labpedia.net).

2.3.2.4 Serum

This is a clear fluid that is separated from the clotted blood. There are no RBCs, White cells, or platelets. There is no need for anticoagulants. Clotted blood is kept at 37 C for at least 20 minutes and then centrifuged; the upper portion is called serum. There is no fibrinogen (Miller., 1984).

Chapter Three
Materials and Methods

Chapter Three

3. Materials and Methods

3.1 Study Design

This was an analytical and a comparative study.

3.2 Study Area

This study was conducted in Khartoum state from April to August 2020.

3.3 Study Population and Sample Size

Sixty Health volunteers randomly selected lived in Khartoum state from different age group both males and females.

3.4 Inclusion Criteria

Health volunteers lived in Khartoum state from different age group both males and females.

3.5 Exclusion Criteria

Individual suffering from any diseases.

3.6 Ethical Consideration

Participants were informed about study, and blood samples were collected after their agreement.

3.7 Sampling Technique

Simple random sampling

3.8 Data Collection

Eight ml of blood were collected from each participant; 2 ml from sample were applied to each container: fluoride oxalate anticoagulant container, sodium citrate anticoagulant container, lithium heparin anticoagulant container and plain container. Then were immediately centrifuged at 3000 rpm for 5 minutes. Samples were examined initially (at zero time), then after 2, 4, 6, and 8 hours from sample collection.

3.9 Methodology

3.9.1 Measurement of glucose:

Serum and plasma glucose were determined by glucose oxidase method.

3.9.2 Principle:

Glucose + O2 + H2O ---glucose oxidase ---- gluconic acid + H2O2

H2O2 + reduced chromogen ---peroxidase---- oxidized chromogen +H2O

Glucose + ATP ----hexokinase----- glucose-6-PO4 + ADP (Bishop, *et al.2018*).

3.9.3Calculations

The glucose concentration in the sample was calculate using the general formula

Sample absorption/ Standard absorption X Standard concentration

(Burtis, *et al*., 2005).

3.9.5 Reference Range

Adult: 74-106 mg/dl or 4.1-5.9 mmol/L (pagana, 2019).

Reagent and procedure refer to appendix page 2

3.10 Quality Control Method

Control sera was used with all samples to ensure the validity of the results.

3.11 Statistical Method

Collecting data were analyzed by using computerized program SPSS.

Chapter Four
Results

Chapter four

4. Results

This study was conducted on 60 healthy adult subject age ranges between 20 to 60 years old. To investigate the stability of blood glucose level in different containers (fluoride oxalate, sodium citrate, lithium heparin and plain container). And different time (at zero time) after 2, 4, 6 and 8 hours from sample collection at room temperature.

The results obtained as in the tables below:

Table (4-1) shows subsequent decrease in glucose concentration in fluoride oxalate stored at room temperature compare with that immediately measured. Which indicate minimal decrease in blood glucose level.

Table (4-2) shows subsequent decrease in glucose concentration in lithium heparin stored at room temperature compare with that immediately measured. Which indicate decrease in blood glucose level in the first 2 hours.

Table (4-3) shows subsequent decrease in glucose concentration in sodium citrate stored at room temperature compare with that immediately measured. Which indicate decrease in blood glucose level in the first 2 hours

Table (4-4) shows subsequent decrease in glucose concentration in serum stored at room temperature compare with that immediately measured. Which indicate decrease in blood glucose level in the first 2 hours.

Table (4-1) Gradual reduction in mean of plasma glucose level at room temperature (25-30 C) at deferent storage time when anticoagulant fluoride oxalate.

Anticoagulants	Time per hours	Mean glucose concentration mg/dl	Reduction of glucose mg/dl	Reduction of glucose %
Fluoride oxalate	Zero time	95.3		
	2 hour	94.1	1.2	1.3
	4 hour	90.4	4.9	5.2
	6 hour	87.4	7.9	8.3
	8 hour	78.9	16.4	17.2

Table (4-2) Gradual reduction in mean of plasma glucose level at room temperature (25-30 C) at deferent storage time when anticoagulant Heparin.

Anticoagulants	Time per hours	Mean glucose concentration mg/dl	Reduction of glucose mg/dl	Reduction of glucose %
Heparin	Zero time	92.8		
	2 hour	89.7	3.1	3.3
	4 hour	83.8	9.0	9.6
	6 hour	73.7	19.1	20.5
	8 hour	65.3	27.5	29.6

Table (4-3) Gradual reduction in mean of plasma glucose level at room temperature (25-30 C) at deferent storage time when anticoagulant Sodium citrate.

Anticoagulants	Time per hours	Mean glucose concentration mg/dl	Reduction of glucose mg/dl	Reduction of glucose %
Sodium citrate	Zero time	91.9		
	2 hour	89.2	2.7	2.9
	4 hour	82.8	9.1	9.9
	6 hour	73.7	17.4	19.8
	8 hour	65.0	25.1	29,2

Table (4-4) Gradual reduction in mean of plasma glucose level at room temperature (25-30 C) at deferent storage time when used plain container.

Anticoagulants	Time per hours	Mean glucose concentration mg/dl	Reduction of glucose mg/dl	Reduction of glucose %
Plain container	Zero time	92.8	0.0	
	2 hour	89.5	3.3	3.5
	4 hour	83.9	8.9	9.5
	6 hour	73.8	19.0	20.4
	8 hour	64.2	28.6	30.8

Table (4-5) comparison of P-value in deferent anticoagulants and plain container at deferent storage time.

P. value				
	Fluoride oxalate	Heparin	Sodium citrate	Plain container
Base line	0.993	0.963	0.632	1
2 hour	0.522	0.093	0.293	0.081
4 hour	0.068	0.000	0.000	0.000
6 hour	0.000	0.000	0.000	0.000
8 hour	0.000	0.000	0.000	0.000

Analyzed by paired T-test.

P-value is significance at 0.005 levels.

Chapter Five

Discussion, Conclusion and Recommendation

Chapter five

5. Discussion, Conclusion and Recommendation

5.1 Discussion:

Glucose is a key to keeping the mechanisms of the body in top working order. This study was conducted on 60 healthy individuals age ranges between 20 to 60 years old to investigate the stability of blood glucose level in different containers (fluoride oxalate, sodium citrate, lithium heparin and plain container) at different time (at zero time 2, 4, 6 and 8 hours) at room temperature

The result of this study showed that blood glucose in fluoride oxalate, heparin and sodium citrate decrease at mean values of 16.4 mg/dl, 27.5 mg/dl and 25.1 mg/dl after 8 hours respectively. with respect to the concentration of glucose before storage, this suggests that storage of blood using fluoride oxalate as an anticoagulant tends to be better preserve the glucose level over a long period of time , this agree with the study done by (Khaled Al salhen., 2017) and another study performed by (Gupta and Kaur., 2014) who agreed and reported that: the ability of fluoride oxalate ion to inhibit the activity of enolase, an enzyme in the glycolytic pathway, thereby slowing down the breakdown of glucose. The random blood glucose significantly ($p \leq 0.05$) decrease steadily as compared to the value before storage. Also (Chan *et al*., 1989) reported that antiglycolytic action of fluoride oxalate is delayed for up to 4 h has little or no effect on the rate of glycolysis during the first 1-2 h after blood is collected.

Serum glucose concentrations with mean value of 28.6 mg/dl. Serum glucose will always be lower than plasma glucose if glycolysis in a plasma sample is inhibited immediately.

The amount of the differences will vary with the glycolysis rate in individual specimens and the time elapsed between collection and centrifugation (Gambino., 2013).

5.2 Conclusion:

The stability of blood glucose is decrease in different anticoagulants with the time increase at room temperature.

The fluoride oxalate is found to be stable for 4 hours, while the other containers; lithium heparin, sodium citrate and plain container cannot save glucose for more than 2 hours. Suggestion that fluoride oxalate is the better.

5.3 Recommendations:

1-The blood sample for glucose should be processed timely after collection.

2- Heparinized plasma, citrated plasma and serum sample for glucose estimation can be estimated with a consideration ratio of an error.

3-More research should be structured in this field.

4-Research on Glucomedics tube for glucose estimation is recommended.

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Appendix (1)

Sudan University of Science and Technology

College of Graduate Studies

Assessment of Glucose Level in Serum and Plasma Using Different Anticoagulants at Room Temperature

Questionnaire

Name……..

Sex: male ( ) female ( )

Age: ……..Years.

Residence:

……

Are you suffering from any of the following diseases?

Diabetes mellitus: ( ) jaundice: ( )

Gout : ( ) renal failure: ( )

Not any one of them: ( )

Are you under treatments?

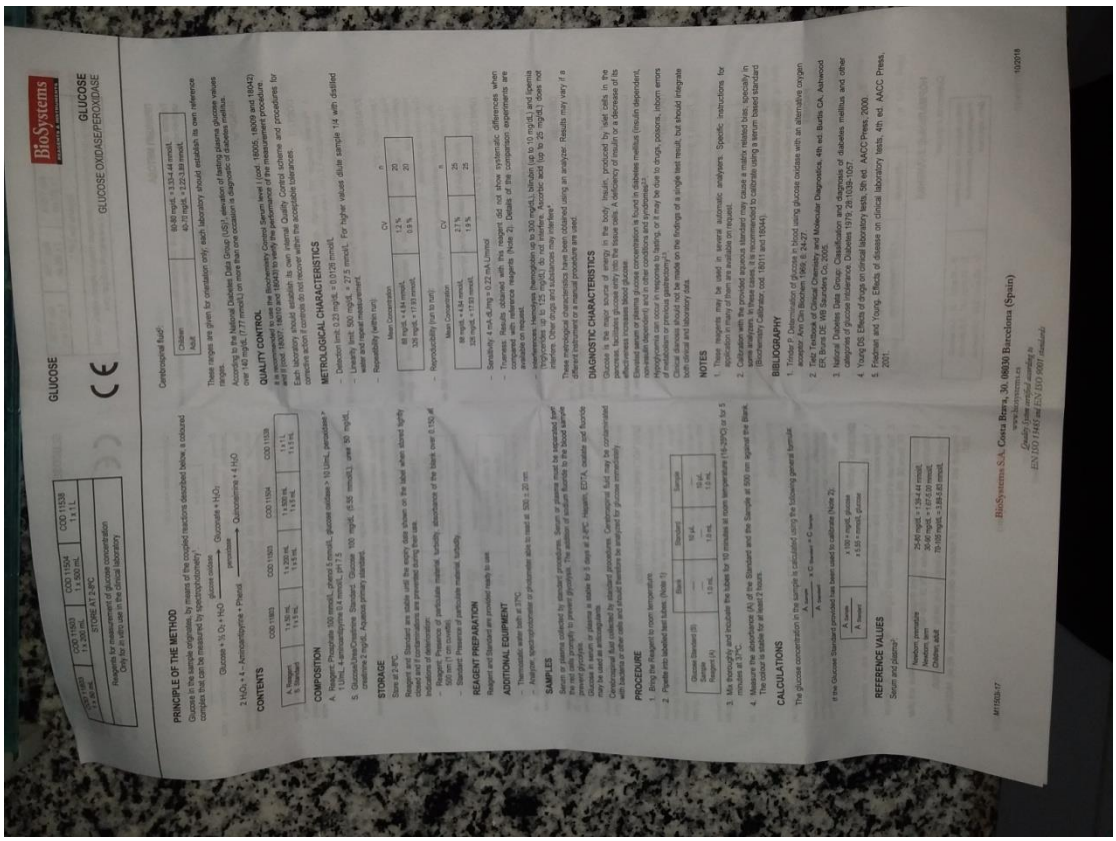
Yes ( ) No ( )

Laboratory Investigations:

These analyzes were performed at room temperature (25 _ 30 c) in serum and plasma.

	Fluoride oxalate	Heparin	Sodium citrate	Plain container
Zero time				
2 hour after collection				
4 hour after collection				
6 hour after collection				
8 hour after collection				

Appendix (2)


BioSystems
GLUCOSE
GLUCOSE
GLUCOSE OXIDASE/PEROXIDASE
PRINCIPLE OF THE METHOD
CONTENTS
COMPOSITION
METROLOGICAL CHARACTERISTICS
QUALITY CONTROL
STORAGE
SAMPLES
REAGENT PREPARATION
ADDITIONAL EQUIPMENT
PROCEDURE
CALCULATIONS
REFERENCE VALUES
DIAGNOSTIC CHARACTERISTICS
NOTES
BIBLIOGRAPHY
BioSystems S.A. Costa Brava, 30. 08030 Barcelona (Spain)
www.biosystems.es
EN ISO 13485 and EN ISO 9001 standards

Appendix (3)