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A COMPARATIVE STUDY OF ERYTHROCYTE SEDIMENTATION RATE (ESR) USING SODIUM CITRATE AND EDTA

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Short Communication

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ABSTRACT

Erythrocyte Sedimentation Rate (ESR) is a marker for inflammation in the body. It is the initial test carried out in order to detect infection using Westergren tube in which blood is mixed with sodium citrate as an anticoagulant. A study was performed using blood from various patients with two different anticoagulants among which one was sodium citrate, commonly used and an alternative to this was Ethylene Diamine Tetra Acetic acid (EDTA). In order to minimise the volume of withdrawal of blood from patients the study of ESR was undertaken with an anticoagulant such as EDTA which is used for Complete Blood Count (CBC) test also. Twenty two blood samples were collected from out- patients. The values of ESR using EDTA showed 4-6 mm less than that of sodium citrate. Thus looking at the results obtained in this comparative study, ESR can be performed using EDTA blood instead of sodium citrate. This utilises only limited amount of blood.

KEYWORDS: Erythrocyte Sedimentation Rate (ESR), EDTA, Sodium citrate, Westergren tube

Introduction

The Erythrocyte Sedimentation Rate (ESR), also known as sedimentation rate or Biernacki Reaction, is the rate at which red blood cells sediment in a period of 1 hour. It is a common haematology test that is a non-specific measure of inflammation. To perform the test, anti-coagulated blood is placed in an upright tube, known as a Westergren tube, and the rate at which the red blood cells fall is measured and reported in millimetres per hour (mm/h).

This test was invented in 1897 by the Polish doctor Edmund Biernacki[1]. In some parts of the world the test continues to be referred to as the Biernacki Test (Polish abbreviation: OB = *OdczynBiernackiego*). In 1918, the Swedish pathologist Robert SannoFåhræus declared the same and along with Alf VilhelmAlbertsson Westergren are eponymously remembered for the Fåhræus-Westergren test (abbreviated as FW test; in the UK, usually termed Westergren test) which uses sodium citrate-coagulated specimens[2,3]. Westergren's normal values, for men 3mm and women 7mm[4]. Normal values are given in **Table 1** as per age and sex.

The ESR is governed by the balance between pro-sedimentation factors, mainly fibrinogen, and those factors resisting sedimentation, namely the negative charge of the erythrocytes (zeta potential). When an inflammatory process is present, the high proportion of fibrinogen in the blood causes red blood cells to stick to each other. The red cells form stacks called 'rouleaux,' which settle faster. Rouleaux formation can also occur in association with some lymphoproliferative disorders in which one or more

immunoglobulins are secreted in high amounts. Rouleaux formation can, however, be a normal physiological finding in horses, cats, and pigs.

Table 1: Normal ESR values with Sodium citrate as an anticoagulant by Westergren Method[5,6,9]

Adults				Children	
Men		**Women**		**Newborn**	**Newborn to puberty**
<50 years	>50 years	<50 years	>50 years	0 to 2 mm/hr	3 to 13 mm/hr
< 15 mm/hr	< 20 mm/hr	< 20 mm/hr	< 30 mm/hr		

Note: mm/hr. = millimetres per hour

Although it can help diagnose some illnesses, an abnormal ESR does not prove that one has a certain condition. Other confirmatory tests are always needed. An increased ESR rate may be due to anaemia, cancers such as lymphoma or multiple myeloma, kidney disease, pregnancy and thyroid disease. An autoimmune disorder is a condition that occurs when the immune system mistakenly attacks and destroys healthy body tissue. ESR is often higher than normal in people with an autoimmune disorder. Common autoimmune disorders include lupus erythematosus and rheumatoid arthritis in adults or children.

Very high ESR levels occur with less common autoimmune disorders including allergic vasculitis, giant cell arthritis, hyperfibrinogenemia, and macroglobulinemia - primary necrotizing vasculitis and polymyalgia rheumatica. An increased ESR rate may be due to some infections including systemic infection, bone infections, infection of the heart or heart valves, rheumatic fever, severe skin infections such as erysipelas and tuberculosis. Lower-than-normal levels occur with congestive heart failure, hypofibrinogenemia, low plasma protein (due to liver or kidney disease), polycythemia and sickle cell anaemia.

Why only sodium citrate is being used as an anticoagulant normally in clinical laboratory for performing ESR? In order to confirm this, ESR was performed using yet another anticoagulant EDTA.

ESR and C-reactive protein (CRP) are both markers of inflammation[5,6]. Generally, ESR does not change as rapidly as does CRP, either at the start of inflammation or as it disappears. CRP is not affected by as many other factors as is ESR, making it a better marker of inflammation. CRP is an acute phase protein produced by the liver during an inflammatory reaction. Since CRP levels in the blood rise more quickly after the inflammatory or infective process begins, ESR is often replaced with C-reactive protein measurement. There are specific drawbacks, however, for example, both tests for ESR and CRP were found to be independently associated with a diagnosis of acute maxillary sinusitis[7] so in some cases the combination of the two measurements may improve diagnostic sensitivity and specificity.

Materials and Methods

This experiment was performed using disposable Westergren tubes. The blood of OPD of Yenepoya University hospital patients were collected into two sets of non-vaccum blood collection vials containing sodium citrate and EDTA respectively. The sodium citrate

non-vaccum blood collection vial contained 3.8% of sodium citrate and EDTA non-vaccum blood collection vial contained 8 mg of EDTA in it. To this 3 ml of blood was added up to the mark and mixed by inverting the tubes, three times. The blood after mixing with anticoagulants was kept for ESR within an hour of collection of blood from the patients. For ESR the non-vaccum blood collection vial are inserted upright with Westergren tube and left as such for 1 hour. The rate of sedimentation of erythrocytes is measured after 1 hour in millimetres. Comparison between ESR of the non-vaccum blood collection vial containing two different anticoagulants is performed and the result is given in mm/hr.

Results and Discussion

As seen from the **Table 2**, the values of ESR using EDTA showed comparatively less by 4-6 mm than that of sodium citrate.

Table 2: Values of ESR with sodium citrate and EDTA blood

Serial No.	Sodium citrate blood (mm/hour)	EDTA blood (mm/hour)
1.	35	30
2.	42	36
3.	56	51
4.	30	24
5.	25	22
6.	38	32
7.	19	13
8.	46	43
9.	128	124
10.	19	19
11.	28	24
12.	30	24
13.	56	51
14.	25	22
15.	38	37
16.	50	45
17.	39	31
18.	89	80
19.	113	107
20.	26	22
21.	45	39
22.	83	78

Although it is frequently recommended, ESR is of limited use as a screening test in asymptomatic patients. It is useful for diagnosing diseases, such as multiple myeloma, polymyalgia rheumatica, various auto-immune diseases, systemic lupus erythematosus, rheumatoid arthritis and chronic kidney diseases. In many of these cases, the ESR may exceed 100 mm/hour[8].

It is commonly used for a differential diagnosis for Kawasaki's disease, an autoimmune disease in which the medium-sized blood vessels throughout the body become inflamed. It may be increased in some chronic infective conditions like tuberculosis and infective endocarditis. It is a component of the PDCAI, an index for assessment of severity of inflammatory bowel disease in children. The use of the ESR as a screening test in asymptomatic persons is limited by its low sensitivity and specificity. When there is a moderate suspicion of disease, the ESR may have some value as a "sickness index."

How can this method be beneficial? While performing ESR another test known as CBC

(Complete Blood Count) is also recommended by the doctor. The CBC test makes use of blood mixed with EDTA as an anticoagulant. So the technicians need to collect blood twice from the patient or they have to take more amount of blood for performing ESR and CBC. As every drop of blood is precious, unnecessary wasting of blood can be minimised, especially in children if ESR is performed by the same blood that is used for CBC. Thus looking at the results obtained in this comparative study, ESR can be performed using EDTA blood instead of sodium citrate. This utilises only limited amount of blood, apart from saving in terms of sodium citrate and additional non-vacuum blood collection vial.

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