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# Research Article

# AUTOMATED ESR ANALYZER: CAN SALINE DILUTED EDTA BLOOD BE USED INSTEAD OF CITRATE DILUTED BLOOD- A COMPARATIVE STUDY

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#### ABSTRACT

Erythrocyte sedimentation rate (ESR) is a simple, cost effective and routinely done hematological test for assessing response to inflammation. Westergrens method of ESR measurement is an error prone and time consuming laboratory test. Semi-automated analyzers have reduced the manual errors and can give the test result in 30 minutes against the 60 minutes taken by the Westergrens method which is the standard test approved by the International Council for Standardization in Hematology. But the drawbacks for this analyzer are that these needed the extra vacutainer. Most hematological tests are run on EDTA blood and citrated blood is used only for ESR. Hence, if EDTA blood could be used for ESR measurement, it would reduce the amount of blood drawn from the patient as well as cut the cost by eliminating the need for citrate vacutainers. Undiluted EDTA blood gives higher values of ESR compared to citrate diluted blood. In this study we would like to assess whether saline diluted EDTA blood could be used as an alternative to citrate diluted blood in the measurement of ESR using automated analyzer. Keywords: Erythrocyte sedimentation rate, citrate, EDTA

#### INTRODUCTION

ESR is a routinely used laboratory test for assessing the inflammatory response.[1] Although it lacks specificity but it can be effective in determining prognosis and monitoring disease activity in any diseases. [2] In the recent years, several newer techniques has been developed introduced in clinical laboratories to address the following needs: (I) to guarantee safety to operators by using automated closed systems; (II) to automate the measurement itself and optimize the workflow and the utilization of human resources at its best; (III) to create unique workstation for measuring ESR and performing other hematologic test (eg, erythrocyte, leukocyte and reticulocyte concentrations) without using multiple specimens. [3]

In recent decades, several new automated ESR analyzer has come, some of these involve automation of the Westergren method with undiluted or diluted samples while other use newer technologies<sup>[4]</sup> like using infrared-ray microphotometer<sup>[3]</sup> and measuring optical density. The benefits of these analyzers are that they are ease of use, economy, practicability, closed sample manipulation and time saving. But these analyzers have to establish their own normal reference ranges and levels of clinical utility, sensitivity and specificity. [4]

In this study we have used the automated analyzer-ESR 2010 which is based on the principle of Infrared technology to measuring ESR. It is using citrate diluted tube blood and gives ESR reading in 30 mins

Most of the hematological tests are done using EDTA blood and citrated blood is used only for ESR. Hence, if EDTA blood could be used for ESR measurement, it would reduce the amount of blood drawn from the patient as well as cut the cost by eliminating the need for citrate vacutainer. [6] According to International committee for standardization in hematology(ICSH) 1993 recommendations both citrate or saline diluted EDTA blood at (4:1) ratio can be used to measure the ESR. [7] So in this study we would like to assess whether saline diluted EDTA blood could be used as an alternative to citrate diluted blood in the measurement of ESR.

# MATERIALS AND METHODS

We have collected 201 samples of patients whom age ranges from 14 to 73 years. Out of which 72 were male and 129 were female. These patients came to both inpatient and outpatient department in RL Jalappa Hospital and Research Centre, Kolar. ESR were measured both by citrated blood and saline diluted EDTA blood in patients who had been advised to get an ESR done.ESR were measured using the ESR 2010 ver 4.0, a semi-automated ESR analyzer. 0.32 ml of 3.8% sodium citrate will be mixed with 1.28 ml whole blood in the citrate vacutainer and 0.32 ml of 0.9 % saline will be mixed with 1.28ml of EDTA anticoagulated whole blood in the EDTA vacutainer. Samples were run in the analyzer within 4 hour of collection of blood. Reading of ESR was taken after 30 mins. A comparison was made between values obtained from these two different anticoagulated blood samples and the results were statistically analyzed using sensitivity, specificity of each test and 95% confidence interval. The receiver operating characteristic (ROC) curve is used to evaluate a diagnostic test.(Figure:1)

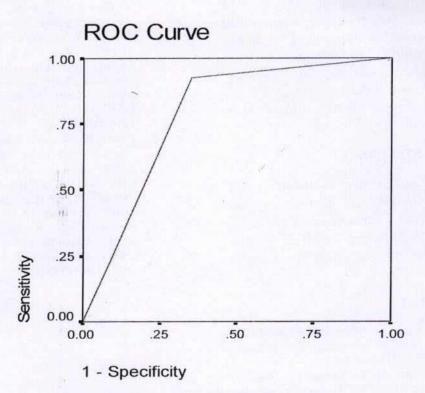
#### RESULTS:

We have collected 201 samples out of which 72 were male and 129 were female. The ESR value got in this study was range from 4 to 128 mm/hour. The sensitivity and specificity of the newer test was 92.86% and 64.71% respectively and their corresponding 95% confidence interval ranges 87.85% to 96.25% and 46.49% to 80.24% respectively. The ROC curve is used in this study to evaluate the performance of this diagnostic test. In our study area depicted under the ROC curve is 0.788. In other words diagnostic test capability of EDTA diluted sample is around 79%. If the area of ROC curve is 75% or more implies test has got good diagnostic valve.

## DISCUSSION:

ESR is although non-specific still but is still used commonly as a indicator of infection and inflammation. [2] The clinical usefulness of ESR is limited such as to monitor the response to therapy in certain inflammatory condition as temporal arteritis, polymyalgia rheumatic and rheumatoid arthritis. [8] It Might denote the presence and severity of particular pathological process. [9] When inflammatory process is present, the high proportion of fibrinogen in the blood causes red cells stick to one another. [10] Ideally trisodium citrate is anticoagulant of choice used to measure the ESR. [8] Some laboratories use EDTA blood dilute with normal saline and other uses whole blood without any diluents to measure the ESR.8 ICSH-1993 recommendations both citrate or saline diluted EDTA blood can be used to measure the ESR. [7]

The blood collected in EDTA enhance the stability, thus favoring rouleaux formation, preserving morphologic features, and precluding nonphysiologic effects on the cells, and consequently being of a great importance for the ESR reaction. [9]



Diagonal segments are produced by ties.

Figure 1: area under the ROC curve is 0.788. Diagnostic test capability will be around 79%

Using the EDTA diluted sample of blood with saline will reduce the cost of the test by using extra citrate vacationer. [5] It not only prevent inconvenient to the patient due to extra sampling technique but also help in withdrawing less blood from the patients. [5]

Earlier manual methods to measure ESR use the principle of sedimentation Westergren pipette or vacuum tube. Automation in measuring the ESR has come in last few decades. [3] Most of these instrument uses citrate diluted blood. [5] These automation help in providing correct dilution of anticoagulant and also the closed system to prevent the exposure of highly infectious blood borne disease like hepatitis B and HIV to the lab personals. [2] The value of these automated analyzer has been compared with gold standard

Westergrens method.<sup>[3]</sup> The ESR 2010 automated analyzer measure ESR using infrared barriers at temperature at 18<sup>o</sup> C.

E Pervovic et al has found that undiluted EDTA sample gave higher values of ESR compared to the citrate diluted sample and also concluded that EDTA can be used to measure ESR in automated analyzer. <sup>[9]</sup> So we have diluted EDTA blood with normal 0.9% saline by ICSH recommendations.

In this study using the EDTA diluted blood sample for measuring ESR we got sensitivity of test around 93% and ROC curve of 79% shows that the test has good diagnostic value.

In conclusion, EDTA diluted sample for determination of ESR is acceptable, accuracy, imprecision and good correlation

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with citrate diluted blood. The use of samples with EDTA as anticoagulant instead of sodium citrate for measuring ESR allows us to do multiple hematological analyses using same anticoagulant, thus avoiding biological hazard, reducing cost of the test and reducing blood sample volume taken from the patient.

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