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Design and Development of a Method to Manipulate Mean Corpuscular Volume of the Red Blood Cells

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Abstract: ***Alsever's solution is a balanced salt solution, is used for preservation of blood. Blood collected from blood bank is treated with low, normal and high alsevers solution to get low, normal and high controls. To this prepare 3.5×10^{12}/L,4.5×10^{12}/L and 5.5×10^{12}/L concentration in 1 ml in EDTA tubes. Then take the values of RBC count and MCV of blood on automated hematology analyser. Alsevers solution can store the blood up to 2 weeks. It is routinely used as an anticoagulant/blood preservative, which permits the storage of whole blood at refrigerator temperatures for approximately 10 weeks. Alsevers solution can give the expected range value for RBC count, MCV, MCH and MCHC.***

Keywords: Alsever's solution, Mean corpuscular volume, Balanced salt solution, Anticoagulant

1. Introduction

Erythrocytes, also known as red blood cells, are biconcave discs that are essential for gaseous exchange. Hemoglobin, which transports oxygen and carbon dioxide throughout the body, is their main product. Each of the four globin subunits that make up hemoglobin surrounds a core hemomoiety. In adults, these subunits are typically 2α and 2β chains. Iron is located in the center of the hemoglobin and is necessary for the movement of gases. The distinctive redness that erythrocytes are known for is produced by hemoglobin. Erythrocytes have an average diameter of 8 μm and a mean volume of 90 fl [1].

Pluripotent hemopoietic cells are used to make red blood cells. These stem cells are found in the bone marrow and eventually mature into fully functional erythrocytes when given the right stimulation from a range of hormones or cytokines red blood cells are specialized in carrying oxygen from pulmonary capillaries to tissue capillaries, where it is exchanged for carbon dioxide. Every minute, a person at rest takes in 250 ml of oxygen and releases 200 ml of carbon dioxide [2] - [3].

The average size of your red blood cells is determined by an MCV blood test. A complete blood count (CBC) often includes an MCV blood test. A routine blood test called a CBC counts various components of your blood, including red blood cells. It is employed to assess your overall well-being. Anemia and other blood disorders can be identified or tracked with the use of an MCV test in conjunction with other diagnostic procedures. Anemia comes in a variety of forms. Finding the right kind of anemia can be aided by an MCV test [4]-[5].

An MCV by itself is insufficient to make a diagnosis. An MCV blood test, when taken into accounts in conjunction with these other tests, can assist your provider in diagnosing conditions and learning more about the condition of your red blood cells [6].

2. Method

2.1 Preparation - Alsever's solutions (1ml)

The packed red blood cells were stored between 2 and 8 °C in a 500 ml glass bottle. This produced three tubes with five milliliters of blood each in a centrifuge tube measuring fifteen milliliters. Centrifuge at 3000 rpm for 5 minutes. Measure the MCV, MCH, and MCHC after discarding the supernatant. List the three control levels for the tubes: low, normal, and high. Resuspend the RBC pellet in LOW, NORMAL, and HIGH Alserver's solution, in increments of 10 milliliters by inversion. Restart the centrifuge at 3000 rpm for five minutes. Delete the supernatant. Repeat the cleaning procedure once more.

In Alsever's solutions, resuspend the RBC suspension in LOW, NORMAL, and HIGH. Examine the MCV, MCH, and MCHC. Prepare RBC concentrations of 3.5×1012/L, 4.5×1012/L, and 5.5×1012/L in 1ml (low, normal, and high) in EDTA tubes using the blood that has been aseptically treated. Read the MCV, MCH andMCHCafter that.

2.2 Calculation for Preparing 1 ml

If the RBC count is (8.5×10^{12}/L)
(a) **Low control (3.5×10^{12}/L)**

8.5×10^{12}/L	-	3.5×10^{12}/L
8.5×10^{9}/ml	-	3.5×10^{9}/ml
8.5/3.5	=	2.428
In 1ml	=	1/2.428
	=	0.411

That means, 411µl blood with 589µl low Alsever's solution is used for the preparation of low control.

(b) Normal control (4.5×10^{12}/L)

8.5×10^{12}/L - 4.5×10^{12}/L
8.5×10^{9}/ml - 4.5×10^{9}/ml
8.5/4.5 = 1.888
In 1ml = 1/1.888
= 0.529

That means ,529µl blood with 471 normal Alsever's solution is used for the preparation of normal control.

(c) **High control ((5.5×10^{12}/L)**

8.5×10^{12}/L - 5.5×10^{12}/L
8.5×10^{9}/ml - 5.5×10^{9}/ml
8.5/5.5 = 1.545
In 1ml = 1/1.545
= 0.647

That means ,647µl blood with 353µl high Alsever's solution is used for the preparation of the high control. Then store the tubes at 4°c. And read the results every day and note the values are in the range of low, normal and high range.
(Low value - ±75,
Normal value - ±85
High values - ± 95).

3. Result

3.1 Changes in MCV with Low, Normal and High Alsever's solution

Solution	*MCV(FL)*
Untreated packed cell	96.1
Low Alsever's solution	82.3
Normal Alsever's solution	95.3
High Alsever's solution	103.1

3.2 Changes of RBC Count and Red Cell Indices in Low, Normal and High Alsever's solution

Solution	*RBCCount (m/cumm)*	*MCV (fl)*	*MCH (pg)*	*MCHC (%)*
Packed RBC	5.13	96.1	30.0	31.2
Alsever's preparation				
Low	4.76	86.2	29.4	34.1
Normal	5.68	91.4	29.0	31.7
High	6.1	103.1	29.8	28.9
1ml preparation				
Low	3.23	83.5	31.2	37.4
Normal	4.46	89.0	28.5	32.0
High	5.54	101.2	29.5	29.1
1st day				
Low	3.42	82.9	29.5	35.6
Normal	4.33	88.3	29.5	33.4
High	5.26	97.4	30.9	31.7
2nd day				
Low	3.42	81.9	29.4	35.9
Normal	4.33	88.1	29.7	33.7
High	5.35	96.7	30.0	31.0
5th day				
Low	3.33	81.5	30.1	36.9
Normal	4.25	87.2	30.0	34.4
High	5.46	96.4	29.3	30.4
10th day				
Low	3.36	80.6	30.1	37.3
Normal	4.20	86.7	30.7	35.4
High	5.54	96.8	29.5	30.5
15th day				
Low	3.71	83.5	26.6	32.0
Normal	4.52	90.2	28.7	31.9
High	5.81	103.8	28.5	27.5
20th day				
Low	3.54	83.3	28.2	34.0
Normal	4.52	87.7	28.7	32.8
High	5.62	89.5	29.3	32.8

The packed red cell count is 5.13×10^{12}/L and means cell volume is 96.1. In the alsever's preparation, the LOW value in the range (3.5×10^{12}/L) and MCV in the range (± 80), NORMAL value in the range(4.5×10^{12}/L) and MCV in the range (±90) and HIGH value in the range (5.5×10^{12}/L) and MCV in the range (±100). In this case, 20 days stability occurs by alsever's preparation.

4. Conclusion

This study highlights the importance of hematological controls in the hematology analyzers. This study helps to prepare red blood cell controls and also help to manipulate mean corpuscular volume of the red blood cells. In this preparation Alsever's solution has an important role to stabilize the red blood cells and mean cell volume. The Alsever's solution stabilizes the red cell volume, so this controls help to use about 20 days at 2-8°c. This preparation gives 20 days open-vial stability at 2-8°c.This preparation has low cost, because expired packed red blood cells are used and also common ingredients are used for the Alsever's preparation. This control preparation is very easy. This type of controls has great benefit in hematology because, it is used in all analyzers. The Alsever's preparation is also used to stabilize the other cells such as WBC and Platelets.

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