



Experimental Studies of Blood Cells and Intensity of Blood Flow

Afshin Zardoshtian¹, Amirreza Rabiee²

¹*Collegium Medicum Nicolaus Copernicus University, Bydgoszcz, Poland*

²*Zanjan University of Medical Sciences, Zanjan, Iran*

Abstract

We have carried out several in vitro experiments for study of erythrocyte aggregation and leukocyte aggregation. After discussing the results, it turned out that it turned out that the change in aggregation forms the blood flow to a greater extent than the aggregation of leukocytes. An increase in aggregation was caused by adding Dextran 500 to the tubes. This is a proven method for increasing aggregation intensity. It turned out that the aggregation of erythrocytes is a stable value compared to the aggregation of leukocytes. It also turned out that the aggregation of erythrocytes changes immediately after the addition of Dextran 500, but not significantly. Research in this direction must continue to receive data how informative the aggregation of erythrocytes and the aggregation of leukocytes are in case of difference diseases.

KEY WORDS: erythrocyte; leukocyte; aggregation; Dextran 500





Introduction

Phenomena such as cold agglutination of red blood cells or platelet aggregation in vitro are well recognized and extensively studied. Nevertheless, aggregation of leukocytes in vitro is a rare phenomenon and may result in pseudoleukopenia. Up until now, the mechanism by which leukocyte aggregation occurs has not been clarified. 3 Two cases of unstable white blood cell counts were detected while we were undertaking leukocyte counts using a hematology analyzer. The 2 different kinds of leukocyte aggregation were identified through microscopic review of stained blood smears.

Aggregation of leukocytes could be due on neutrophil aggregation, aggregation of lymphocyte, leukocyte-platelet aggregation. Last position might relate to malignancies, infections, hepatic disorders, or autoimmune diseases. This could be in patients' group with had an acute infection and also without obvious underlying disease.

There is very interesting article about the mechanism of leukocyte aggregation is mainly related to the use of K2 EDTA anticoagulant or is temperature-mediated. IgM can increase obviously and the neutrophils aggregated in a short time. Therefore, the aggregation can dependent on temperature. This indicated that a form of K2 EDTA-related and temperature-dependent antineutrophil IgM antibody may exist in the serum.

There the possible existence of a type of K2 EDTA-related anti-leukocyte IgM antibody in the serum [1].

Studying rheology, begin to understand that, rheology is a very significant clinical science for measure of blood flow. Rheology unites the doctrine of the flow of biological fluids such as blood, plasma, cerebrospinal fluid, etc. Hemorheology mainly investigates blood flow through the vessels in the macro and microcirculation [2,3]. The basic concepts of hemorheology are the aggregation ability of blood cells. Until recently, the main emphasis was on the erythrocytes aggregability. In the last time, researchers have started to show particular interest in the study of the aggregation ability of leukocytes [4].

The purpose of our work was to study the aggregation potential of erythrocytes in blood samples from healthy people after the addition of Dextran 500. After 1 hour, after 2 hours, after 8 hours. Dextran 500 is known as an aggregation enhancer [5]. However, our interest was directed to how much the increase in erythrocytes aggregation and leukocytes aggregation leukocytes is persistent. However, our interest was directed to how much the increase in erythrocytes aggregation and leukocytes aggregation are persistent.





Material and Methods

By the technology could establish the leukocytes differential using 2 measurements: individual cell volume, high-frequency conductivity. Would be better, if we can study of laser-light scatter. But in the goal of study this stage was not. The leukocytes' volume is measured using impedance; the cells were in their near-native state, and this gives a good indication of the cell volume as it circulates in the blood [6, 7]. Age of volunteers (man) was $24 \pm 3,5$ years. $n=6$ (I, II, III, IV, V, VI cases). Aggregation of erythrocytes counts as the area of aggregated erythrocytes divided by the full area of the erythrocytes in volume unit. This can be done with the textural analyses system. This new innovative method is famous in the world as direct, numeral and exact [5]. Our research protocol looked like this. That was an in vitro study. In a drop of blood from healthy volunteers, we examined the number of erythrocytes and the number of leukocytes, as well as the erythrocytes aggregability and leukocytes aggregation.

Results

We measured the number of leukocytes and erythrocytes, the aggregation of erythrocytes and leukocytes in a test tube with the blood of healthy people. Then the aggregation of erythrocytes and leukocytes after 1, 2, 8 and 24 hours was measured in the same test tubes after the addition of Dextran 500. See table for measurement results.





Table 1. The number of leukocytes and erythrocytes, the aggregation of erythrocytes and leukocytes in a test tube with the blood of healthy people. Then the aggregation of erythrocytes and leukocytes after 1, 2, 8 and 24 hours was measured in the same test tubes after the addition of Dextran 500. $n=6$.

Parameters	I case	II case	III case	IV case	V case	VI case
RBC $10^{12}/l$	4,2	4,5	5,2	4,9	4,3	5,0
WBC $10^9/l$	8,2	6,0	7,8	7,0	6,5	7,9
RBCAI before	15	16	16	13	15	17
WBCAI before	1,8	1,9	1,7	1,9	1,5	1,9
RBCAI after 1 hour	25	32	24	35	30	27
WBCAI after 1 hour	2,5	2,4	2,5	2,3	2,5	2,4
RBCAI after 2 hours	26	34	30	45	36	35
WBCAI after 2 hours	2,4	2,3	2,2	2,2	2,2	2,4
RBCAI after 8 hours	37	48	50	57	48	49
WBCAI after 8 hours	1,8	2,0	1,7	1,7	1,5	1,8
RBCAI after 24 hours	stasis	stasis	stasis	stasis	stasis	stasis
WBCAI after 24 hours	1,7	1,8	1,7	1,8	1,5	1,8

Discussion

When measuring the aggregation of erythrocytes and aggregation of leukocytes, it turned out that the change in aggregation forms the blood flow to a greater extent than the aggregation of leukocytes. An increase in aggregation was caused by adding Dextran 500 to the tubes. This is a proven method for increasing aggregation intensity. It turned out that the aggregation of erythrocytes is a stable value compared to the aggregation of leukocytes. It also turned out that the aggregation of erythrocytes changes immediately after the addition of Dextran 500, but not significantly. This change was within normal limits. In this case, the aggregation of leukocytes also changes, but is restored after 2 hours, while the



aggregation of erythrocytes increases all the time and after 24 hours the blood in the test tube stasis, while the leukocyte aggregation is restored.

Despite the fact that we examined only 6 cases, the results emphasized that the aggregation of erythrocytes is a value that reacts late to the pathogen (in our case, Dextran 500), but cannot be restored, while the aggregation of erythrocytes is restored after a certain time. Our research started a joint research with Prof. Mantskava (Georgia). In the framework of our new study, we will be able to conclude how informative the aggregation of erythrocytes and the aggregation of leukocytes are in case of infarction and arterial hypertension.

Describe of shortening

[RBC] – erythrocyte

[WBC] – leukocyte

[RBCAI] – erythrocyte aggregation index

[WBCAI] – leukocyte aggregation index

References

1. Dagan Yang, MS, Xichao Guo, BS, Yu Chen, MD, Genyun Xu, BS. Leukocyte Aggregation in vitro as a Cause of Pseudoleukopenia, Laboratory Medicine. 2008; 39(2):89-91. <https://doi.org/10.1309/RHN7D-0FRW6WOQKWR>
2. Wilson DI. What is rheology? Eye (Lond). 2018; 32(2):179-183. doi: 10.1038/eye.2017.267. Epub 2017 Dec 22. PMID: 29271417; PMCID: PMC5811736.
3. Zhang JX, Feng Y, Zhang Y, Liu Y, Li SD, Yang MH. Hemorheology Index Changes in a Rat Acute Blood Stasis Model: A Systematic Review and Meta-Analysis. Afr J Tradit Complement Altern Med. 2017; 14(4):96-107. doi: 10.21010/ajtcam.v14i4.12. PMID: 28638872; PMCID: PMC5471488.
4. Kang YJ. Microfluidic-Based Measurement Method of Red Blood Cell





- Aggregation under Hematocrit Variations. *Sensors (Basel)*. 2017; 17(9):2037. doi: 10.3390/s17092037. PMID: 28878199; PMCID: PMC5620946.
5. Abir F, Barkhordarian S, Sumpio BE. Efficacy of dextran solutions in vascular surgery. *Vasc Endovascular Surg*. 2004; 38(6):483-91. doi: 10.1177/153857440403800601. PMID: 15592628.
 6. Mchedlishvili G. Hemorheological changes in microcirculation: their mechanism and measurement technique. *Indian J Exp Biol*. 2007; 45(1):32-40. PMID: 17249325.
 7. Mantskava M, Momtselidze N. Erythrocytes aggregability and deformability in residual umbilical cord blood *Series on Biomechanics* 2018; 32(2):24-27 http://jsb.imbm.bas.bg/page/en/details.php?article_id=279

