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Ability of Erythrocytes to Absorb Different Endogenous Substances

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Abstract In order to study the sorption capacity of erythrocytes in erythrocyte masses prepared in different forms, erythrocyte mass was isolated from the blood of 48 male donors of Uzbek nationality aged 21-62 at the Republican Blood Transfusion Center. leukofiltered. The ability of erythrocytes to absorb endogenous substances such as total protein, albumin, glucose, cholesterol, triglyceride, high-density lipoprotein (HDLP), low-density lipoprotein (LDLP), urea and creatinine was studied. In the obtained results, there are significant changes in the ability of sorption of various endogenous substances in washed erythrocytes. In this case, the rate of sorption of endogenous substances mainly increases. In irradiated erythrocytes, there are significant changes in the ability to absorb various endogenous substances. In this case, not only sorption of endogenous substances, but also reverse processes, i.e. desorption, are observed. In frozen erythrocytes, there are significant changes in the ability to absorb various endogenous substances. In this case, the ability to sorb endogenous substances is observed for all substances, and for triglycerides, the reverse process, desorption, is also observed. In leukofiltered erythrocytes, there are significant changes in the ability to absorb various endogenous substances. In this case, erythrocytes almost lose their ability to absorb many endogenous substances.

Keywords Donor, Blood, Erythrocyte, Sorption, Desorption

1. Introduction

The predominance of the adsorption of one substance can disrupt the metabolism of others, thereby creating the necessary conditions for hidden pathologies that cannot be detected in blood plasma analysis. The practical importance of the adsorption-transport function of erythrocytes has been considered in relation to diabetes and atherosclerosis [1].

Adsorption of proteins in erythrocytes affects the deformation and rheology of blood, and the adsorbed part of the protein is a reserve for emergency replenishment of proteins in the plasma [2]. It was found that the process of transporting organic substances in erythrocytes is more variable and demonstrative compared to the corresponding plasma indicators. It has been proven to regulate transport of substances in erythrocytes and maintain adsorption due to the activity of physicochemical bonds of hemoglobin inside erythrocytes [3]. Thus, there is a second important function of erythrocytes - adsorption-transport function, which plays an important role in the processes of rapid and selective entry of substances into the exchange layer of blood capillaries [4]. The passage of erythrocytes through the narrow parts of the capillaries ensures mechanical replacement of molecules adsorbed on erythrocytes with substances in the wall exchange layer. Differences in the ability of substances to be adsorbed on the surface of erythrocytes made it possible to divide them into weakly, moderately and strongly adsorbed types.

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2. Purpose of the Research

The goal of our scientific work is to study the sorption capacity of erythrocytes in erythrocyte masses prepared in different forms.

3. Material and Methods

Due to the fact that the research was conducted only on the blood of donors, blood was taken from a total of 48 male donors of Uzbek nationality aged 21-62 at the Republican Blood Transfusion Center (Tashkent, Republic of Uzbekistan). Blood donors are from almost all regions of the republic, and among them there are both urban and rural residents. Donors were informed that their biological material would be used in the intended research study and their consent was obtained.

In the general analysis of the blood of all studied donors, the following were determined: the amount of erythrocytes (RBC), leukocytes (WBC), platelets (PLT) and hemoglobin (Hb). The erythrocyte mass was separated from the collected blood, washed erythrocytes were obtained from one part, one part was frozen, one part was irradiated, and another part was leukofiltered. These procedures were carried out together with specialists of the Republican Blood Transfusion Center. The ability of erythrocytes to absorb endogenous substances such as total protein, albumin, glucose, cholesterol, triglyceride, high-density lipoprotein, low-density lipoprotein, urea and creatinine was studied. The content of blood plasma was determined using an automatic biochemical analyzer HumaStar 100 (Federal Republic of Germany).

To determine the rate of absorption of erythrocytes, the blood collected in a test tube with heparin was centrifuged at 3000 rpm for 10 minutes, and the plasma was separated.

Work progress:

1. Total protein (g/l), albumin (g/l), glucose (mmol/l), cholesterol (mmol/l), triglycerides (mmol/l) in the isolated plasma using the automatic biochemical analyzer HumaStar 100 (Federal Republic of Germany), urea (mmol/l) and creatinine (μmol/l) were determined.
2. The erythrocyte masses were mixed with plasma in a ratio of 1:1, mixed carefully and left at room temperature for 10 minutes.
3. Then the tubes were again centrifuged at 3000 rpm. Plasma was isolated and the above endogenous substances were determined again in a biochemical analyzer.
4. To determine the rate of absorption of erythrocytes, we divided the difference of results 1 and 2 by 10 and found the change in concentration in 1 minute.

4. Results and Discussion

Our research to study the sorption capacity of erythrocytes in erythrocyte masses prepared in different forms showed that the sorption capacity of washed erythrocytes was 3 times higher than the control (Table 1). That is, control erythrocytes adsorbed 489 mg of protein per 1 minute, while washed erythrocytes adsorbed 1440 mg of protein per 1 minute. The albumin sorption capacity of the washed erythrocytes was also 1.3 times higher than the control (900 mg/min versus 711 mg/min in the control). Glucose uptake in controls was 148 μmol/min, whereas washed erythrocytes absorbed 838 μmol/min of glucose.

We also studied the ability of washed erythrocytes to absorb lipids. The results of the study showed that the cholesterol sorption capacity of the washed erythrocytes increased by 2.7 times compared to the control. The ability of washed erythrocytes to absorb triglycerides was 1.5 times higher.

The rate of sorption of HDLP by control and washed erythrocytes did not differ from each other. At the same time, the sorption of LDLP by washed erythrocytes was 1.7 times lower than the control. The urea absorption rate of washed erythrocytes was 1.2 times higher than the control, and the creatinine sorption capacity was 4.9 times higher.

The erythrocyte sorption capacity was also studied in irradiated erythrocytes. The results of the study showed that the ability of irradiated erythrocytes to absorb total proteins was 3.7 times higher than that of the control. While control erythrocytes adsorbed 489 mg of protein per 1 minute, irradiated erythrocytes adsorbed 1560 mg of protein per 1 minute.

No change in albumin sorption capacity of irradiated erythrocytes was observed. Glucose absorption of irradiated erythrocytes was 100%. The study of lipid sorption ability of irradiated erythrocytes showed that when they were incubated with primary plasma, the amount of cholesterol in the incubation medium increased by 14%. This means that instead of sorption, desorption has gone. Calculations showed that irradiated erythrocytes desorbed cholesterol into the medium at a rate of 24 μmol per minute.

Irradiated erythrocytes almost did not absorb triglycerides and HDLP. At the same time, desorption of LDLP was observed in irradiated erythrocytes.

In this case, these lipoproteins were released from erythrocyte membranes into the environment in the amount of 28 μmol per minute. Irradiated erythrocytes also desorbed urea, where the desorption rate was 102 μmol/min (sorption was observed in the control). Irradiated erythrocytes hardly absorbed creatinine.

When studying the sorption capacity of frozen erythrocytes, it was found that their total protein sorption capacity was 4.2 times higher than that of the control. While control erythrocytes adsorbed 489 mg of protein per 1 minute, frozen erythrocytes adsorbed 2060 mg of protein per 1 minute. Frozen erythrocytes also actively sorbed albumin. In this case, the control erythrocytes absorbed 711 mg of albumin per minute, while the frozen erythrocytes absorbed 1600 mg of albumin per minute, that is, the sorption capacity of frozen erythrocytes increased by 2.3 times (230%) compared to the control. Glucose absorption of frozen erythrocytes was 4.3 times higher than that of control, that is, control erythrocytes absorbed 148 μmol of glucose per minute, while frozen erythrocytes absorbed 630 μmol of glucose per minute. The study of the lipid sorption capacity of frozen erythrocytes showed that when they were incubated with primary plasma, the amount of cholesterol in the incubation medium was reduced by 35.1%.

In this case, frozen erythrocytes absorbed 61 μmol of cholesterol per minute instead of 36 μmol of cholesterol in the control. Frozen erythrocytes desorb triglycerides. While control erythrocytes sorbed 15 μmol of triglycerides per minute, frozen erythrocytes desorbed 223 μmol of triglycerides per minute into the medium. HDLPs and LDLP were 100% sorbed by frozen erythrocytes. In this case, the sorption rate of HDLPs was 4.2 times higher than the control, and LDLP was 2.8 times higher. The frozen erythrocyte

urea uptake rate was 1.8 times higher than the control, and creatine sorption was 6.0 times higher. In this case, the urea uptake of frozen erythrocytes was 140 μmol of urea per minute instead of 78 μmol of the control. The rate of creatinine sorption by frozen erythrocytes was 3880 nmol per minute instead of 644 nmol per minute in the control.

Leukofiltered erythrocytes did not absorb total proteins. In this case, the sorption of 20 mg/min was not statistically reliable. The same situation was observed in the sorption of albumin (the sorption of 140 mg per minute was also not statistically reliable). Glucose uptake of leukofiltered erythrocytes was 100% of that of irradiated erythrocytes, and the glucose uptake rate of these erythrocytes was 2210 μmol/min (148 μmol in control).

Table 1. The ability of erythrocytes in the erythrocyte mass to absorb various substances from the blood plasma

Indicators, dimensionality	Statistical indicators	Erythrocytes				
		Control	Washed	Illuminated	Frozen	Filtered
Total protein, g/l	M ± m, until incubation	68,22±1,04	48,0±2,49[a]	58,0±2,02[a]	60,0±1,46[a]	60,0±1,05[a]
	M ± m, after incubation	63,33±1,76[b]	33,6±1,35[a,b]	42,4±1,08[a,b]	39,4±1,46[a,b]	59,8±1,50
	Decrease (-), increase (+), %	- 7,2	- 30,0	- 26,9	- 34,3	- 0,3
	Sorption, desorption (*), mg/min	489	1440	1560	2060	20
albumin, g/l	M ± m, until incubation	52,78±1,19	32,0±1,39[a]	42,0±1,33[a]	44,0±1,10[a]	44,00±1,30[a]
	M ± m, after incubation	45,67±1,37[b]	23,00±0,45[a,b]	42,4±1,08	28,0±1,07[a,b]	42,6±1,00[a]
	Decrease (-), increase (+), %	- 13,5	- 28,1	+ 0,95	- 36,4	- 3,2
	Sorption, desorption (*), mg/min	711	900	40*	1600	140
Glucose, mmol/l	M ± m, until incubation	4,89±0,11	22,1±1,19[a]	22,1±1,0[a]	22,1±0,93[a]	22,1±0,54[a]
	M ± m, after incubation	3,41±0,13[b]	13,72±0,22[a,b]	0	15,8±0,27[a,b]	0
	Decrease (-), increase (+), %	- 30,3	- 37,9	- 100	- 28,5	- 100
	Sorption, desorption (*), μmol/min	148	838	2210	630	2210
Cholesterol, mmol/l	M ± m, until incubation	5,34±0,22	1,74±0,11[a]	1,71±0,06[a]	1,74±0,06[a]	1,74±0,05[a]
	M ± m, after incubation	4,98±0,23	0,76±0,06[a,6]	1,95±0,03[a,b]	1,13±0,04[a,b]	2,05±0,06[a,b]
	Decrease (-), increase (+), %	- 6,7	- 56,3	+ 14,0	- 35,1	+ 17,8
	Sorption, desorption (*), μmol/min	36	98	24*	61	31*
Triglycerides, mmol/l	M ± m, until incubation	1,40±0,17	0,80±0,03[a]	1,00±0,03[a]	0,97±0,02[a]	0,97±0,02[a]
	M ± m, after incubation	1,25±0,19	0,58±0,01[a,b]	0,98±0,03	3,2±0,89[a,b]	1,02±0,03
	Decrease (-), increase (+), %	- 10,8	- 27,5	- 2	+ 229,9	+ 5,2
	Sorption, desorption (*), μmol/min	15	22	2	223*	5*
HDLP, mmol/l	M ± m, until incubation	1,15±0,05	0,46±0,04[a]	0,59±0,02[a]	0,59±0,03[a]	-
	M ± m, after incubation	1,01±0,07	0,31±0,02[a,b]	0,62±0,02[a]	0	-
	Decrease (-), increase (+), %	- 12,2	- 32,6	+ 5,1	- 100	-
	Sorption, desorption (*), μmol/min	14	15	3*	59	-
PZLP, mmol/l	M ± m, until incubation	3,65±0,20	0,71±0,03[a]	0,91±0,06[a]	0,91±0,05[a]	-
	M ± m, after incubation	3,33±0,21	0,52±0,01[a,b]	1,19±0,02[a,b]	0	-
	Decrease (-), increase (+), %	- 8,8	- 26,8	+ 30,8	- 100	-
	Sorption, desorption (*), μmol/min	32	19	28*	91	-
Urea, mmol/l	M ± m, until incubation	5,44±0,35	3,10±0,17[a]	3,30±0,13[a]	3,70±0,11[a]	3,70±0,10[a]
	M ± m, after incubation	4,66±0,32	2,18±0,15[a,b]	4,32±0,14[b]	2,3±0,10[a,b]	3,80±,010[a]
	Decrease (-), increase (+), %	- 13,3	- 29,7	+ 30,9	- 37,8	+ 2,7
	Sorption, desorption (*), μmol/min	78	92	102*	140	10*
Creatinine, μmol/l	M ± m, until incubation	104,22±5,17	135,0±3,49[a]	151,0±3,10[a]	151,0±2,73[a]	151,0±1,82[a]
	M ± m, after incubation	97,78±5,46	103,40±1,77[b]	157,6±2,79[a]	112,2±2,74[a,b]	149,0±1,84[a]
	Decrease (-), increase (+), %	- 6,2	- 23,4	+ 4,4	- 25,7	- 1,3
	Sorption, desorption (*), nmol/min	644	3160	660*	3880	200

Note: *a – reliable difference compared to control; b - reliable difference in relation to incubation; * - desorption*

Cholesterol desorption was observed in leukofiltered erythrocytes, as well as in irradiated erythrocytes. In this case, leukofiltered erythrocytes desorbed 31 μmol of cholesterol per minute instead of 36 μmol of cholesterol in the control. Leukofiltered erythrocytes did not absorb triglycerides. In this case, the observed desorption of 5 μmol/min was not statistically reliable. HDLPs and LDLP were not detected in the primary medium of leukofiltered erythrocytes. No changes were observed in urea uptake rate and creatinine sorption rate of leukofiltered erythrocytes, that is, urea desorption of 10 μmol/min and creatinine sorption of 200 nmol/min of leukofiltered erythrocytes were not statistically reliable.

5. Conclusions

The obtained results indicate that certain changes occur in the cells during the preparation of erythrocyte-preserving drugs. These results lead to the conclusion that in the future, the use of erythrocyte mass erythrocytes prepared by different methods should be approached taking into account their ability to absorb substances.

Based on the analysis of the obtained results, we present the following conclusions:

1. In the washed erythrocytes, significant changes in the ability to absorb various endogenous substances are observed. In this case, the rate of sorption of endogenous substances mainly increases.
2. In irradiated erythrocytes, there are significant changes in the ability to absorb various endogenous substances. In this case, not only sorption of endogenous substances, but also reverse processes, i.e. desorption, are observed.
3. Big changes are also observed in frozen erythrocytes in their ability to absorb various endogenous substances. In this case, the ability to sorb endogenous substances is observed for all substances, and for triglycerides, the reverse process, i.e., desorption, is also observed.
4. In leukofiltered erythrocytes, great changes are observed in the ability to absorb various endogenous substances. In this case, erythrocytes almost lose their ability to absorb many endogenous substances.

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