# RATE OF POTASSIUM EXCHANGE OF THE HUMAN ERYTHROCYTE*

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When the radioactive isotope of potassium, $K^{42}$, became available, it was possible to observe the normal exchange of potassium between the intracellular phase of erythrocytes and their extracellular medium. The *in vivo* work of Hahn, Hevesy, and Rebbe (1) in 1939 and of Cohn (2) in 1941 showed that some exchange of potassium does occur in mammalian erythrocytes. Dean and Fenn and their collaborators (3, 4) showed that the red cell membrane was permeable *in vitro* and *in vivo* to potassium and calculated exchange rates.

The following experiments were undertaken to determine the rate of transfer of potassium across the human erythrocyte membrane in a stable *in vitro* system. For this purpose human red cells were incubated in diluted plasma containing radioactive potassium as a tracer. The change with time of the specific activity of the extracellular potassium was determined and the rate of transfer of potassium between the intracellular and extracellular phases was calculated.

## *Methods*

The rocker-dilution technique developed by Geiman *et al.* (5) for the cultivation of malarial parasites was used. This permits the incubation of red cells for 52 hours with minimal change in pH. Centrifugation, washing, and excessive agitation of the red cells prior to incubation were avoided because of the demonstrated effects of mechanical procedures on the permeability of red cells (6).

Two parts of freshly drawn, heparinized, whole blood were diluted with three parts of an inorganic medium having the composition shown in Table I. This medium has a potassium concentration of 4.42 millimoles per liter and a freezing point of $-0.56°C$. About 1 millicurie of radioactive potassium was present per liter of medium. When in later experiments the amount of potassium chloride was changed, the sodium chloride was changed by an equivalent amount in order to keep the medium isotonic. Glass

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redistilled water was used for making all solutions. Glucose was added to make the initial concentration from 2.5 to 5.0 mg. per ml. No other organic constituents were added. The medium was equilibrated with 5 per cent carbon dioxide before use to bring the pH to 7.4.

In a typical experiment, 80 ml. of adult male human venous blood was drawn in a sterile syringe containing 2 ml. of heparin (liquaemin). This was then introduced into a flask containing 120 ml. of sterile medium and mixed by rotation. The time of mixing was recorded as zero time and an aliquot was withdrawn for the determinations described below. The remainder of the mixture was placed in sterile rocker-dilution vessels (about 12 ml. in each). These were rocked 12 times a minute in a constant temperature room. For the 18 experiments in which the normal exchange rate was determined, the temperatures of this room were between 34° and 39°C. During any one experiment the temperature varied ±0.5°C. The mixture was equilibrated

TABLE I

*Composition of Diluting Medium*

| | Amount | Amount |
|---|---|---|
| | *gm./l.* | *mM/l.* |
| $MgCl_2 \cdot 6H_2O$ | 0.203 | 1.00 |
| $CaCl_2$ | 0.056 | 0.50 |
| $Na_2HPO_4$ | 0.242 | 1.70 |
| $NaHCO_3$* | 2.350 | 27.97 |
| $KH_2PO_4$ | 0.057 | 0.42 |
| NaCl | 5.913 | 101.16 |
| KCl + $K^{42}Cl$ | 0.298 | 4.00 |

* Added as $Na_2CO_3$ (1.480 gm./liter) and converted to $NaHCO_3$ by passing 100 per cent $CO_2$ through the solution.

during incubation with 5 per cent carbon dioxide in air or oxygen. The duration of the experiments ranged from 24 to 52 hours. At intervals aliquots of the incubating mixture were withdrawn under sterile conditions for the following determinations: hematocrit, diluted plasma potassium concentration, diluted plasma radioactivity, diluted whole blood potassium concentration, and diluted whole blood radioactivity. In some experiments glucose, plasma pH, and lactate determinations were also done.

Specifically, a sample of the diluted whole blood mixture was transferred to a graduated hematocrit tube of 2.5 ml. capacity (dimensions, 15 × 0.5 cm.). This was centrifuged for 30 minutes at 1800 R.P.M. at a radial distance of 25 cm. After reading the percentage of cells of the diluted whole blood mixture, the diluted plasma was pipetted off and used for potassium and radioactivity determinations.

All potassium concentrations were determined on the Perkin-Elmer flame photometer model 52A using as an internal standard lithium sulfate monohydrate (Baker). For potassium determinations on diluted plasma, 1.0 ml. of diluted plasma plus 1.0 ml. of lithium sulfate (150 m. eq. per liter) were diluted to 25.0 ml. with glass redistilled water; for those on diluted whole blood, 1.0 ml. of blood mixture plus 4.0 ml.

of lithium sulfate solution were diluted to 100.0 ml. with water. The final potassium concentrations were between 0.15 and 0.25 m. eq. per liter, and the final lithium concentration was 6 m. eq. per liter. All determinations were made in triplicate and four readings were recorded for each solution. The error in the determination of plasma potassium was less than 2 per cent as checked by recovery experiments and chemical determinations.

For the determination of radioactivity 0.2 ml. samples of diluted plasma and whole blood were plated in aluminum cups having a 2.0 cm. diameter. Lens paper was placed in the bottom of each cup to aid in even distribution. The samples were dried at 37°C. All radioactivity determinations were recorded as counts per minute per 0.2 ml. sample. The error in counts between duplicate samples was less than 5 per cent.

All glucose determinations were done by the Nelson-Somogyi method (7).

Lactate determinations were done by the method of Barker and Summerson (8).

pH determinations were done by the colorimetric method of Hastings and Sendroy (9).

### *Stability of the System*

In order to treat the data in the mathematical manner to be described in the next section, it is necessary that the system under investigation be essentially stable with respect to the distribution of potassium between the intracellular and extracellular phases. Moreover, for the investigation of red cell ionic exchange, it is desirable that the metabolism of the cell be proceeding in as normal a fashion as possible. The data in this section are presented for the evaluation of our system on the basis of these criteria.

During 48 hours of incubation, there occurs an increase in the amount of intracellular potassium at the expense of the extracellular potassium. This has been previously reported by Danowski (10). The total change is about 3 per cent of the erythrocyte potassium. The erythrocytes also slowly take on water as evidenced by an increase in the hematocrit determination. These changes for two experiments are shown in Table II. The calculated ratio of the intracellular potassium concentration per liter of cell water to the plasma potassium concentration per liter of plasma water is based on the assumptions that the initial cell water is 65 per cent and the initial diluted plasma water is 97 per cent. The concentration gradient between the intracellular and extracellular potassium when calculated on this basis is essentially unaltered in one experiment over a 48 hour period and was unchanged in the other experiment for a 24 hour period. After 48 hours of incubation, no hemolysis is visible in the system, and the red cells appear microscopically normal. The plasma pH at the start is 7.4 to 7.5 and falls to 7.2 to 7.3 at 24 hours and to 7.0 to 7.1 at 48 hours. Glucose utilization in our experimental system is at the rate of about 230 mg. per liter of red cells per hour and the rate of lactate production is about 160 mg. per liter of cells per hour. If the glucose is exhausted, the cells lose potassium to the external fluid (10). Initial glucose concentrations were high enough to insure that the glucose was not completely utilized during incubation.

*Calculation of the Exchange Rate*

The specific activities of the diluted plasma potassium and of the red cell potassium plotted against time for one experiment are shown in Fig. 1. When

TABLE II

*Distribution of Potassium between Diluted Plasma and Cells during Incubation at 37°C.*

| Time | $[K^+]_p$/l. plasma | Hematocrit determination | $[K^+]_p$/l. blood | $[K^+]_c$/l. blood | Change in $[K^+]_c$ | $\frac{[K^+]_c/l.H_2O_c}{[K^+]_p/l.H_2O_p}$ |
|---|---|---|---|---|---|---|
| *hrs.* | *mM/l.* | *per cent* | *mM/l.* | *mM/l.* | *per cent* | |
| 0.3 | 4.37 | 17.5 | 3.61 | 16.97 | 0 | 33.01 |
| 13.0 | 4.12 | 17.6 | 3.39 | 17.19 | +1.3 | 35.25 |
| 22.6 | 3.95 | 18.3 | 3.23 | 17.35 | +2.2 | 34.85 |
| 39.1 | 3.87 | 19.2 | 3.13 | 17.45 | +2.8 | 33.30 |
| 47.2 | 3.84 | 19.7 | 3.08 | 17.50 | +3.1 | 32.50 |
| 0.3 | 4.17 | 19.5 | 3.36 | 18.19 | 0 | 31.54 |
| 12.9 | 3.91 | 20.1 | 3.12 | 18.43 | +1.3 | 32.61 |
| 24.9 | 3.78 | 21.1 | 2.98 | 18.57 | +2.0 | 31.53 |
| 35.5 | 3.86 | 21.7 | 3.02 | 18.53 | +1.9 | 29.55 |
| 47.9 | 4.15 | 22.4 | 3.22 | 18.33 | +0.8 | 25.94 |

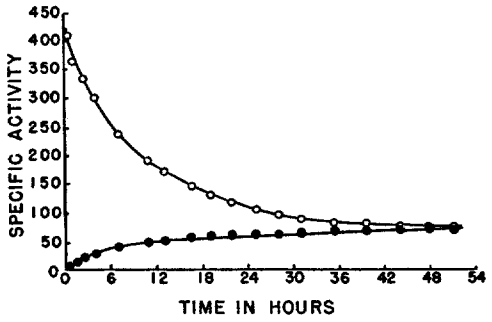


FIG. 1. The specific activities of the diluted plasma potassium and of the calculated cell potassium plotted against time. ○ represents diluted plasma potassium, and ● represents cell potassium.

equilibrium is attained, the ratio of the specific activity of the plasma potassium to the specific activity of the red cell potassium would be 1.0. At 50 hours this value is reached within an error of 2 per cent. This is evidence that all the red cell potassium is exchangeable.

If the sytem under discussion be considered to be two opposing unimolecular reactions, it is possible to calculate rate constants expressing the fraction of diluted plasma potassium and the fraction of erythrocyte potassium transferred across the membrane per unit time. No implications as to the mechanisms of this transfer are inherent in these calculations.

In the opposing reactions $A \rightleftarrows B$, if $x$ moles of $A \rightarrow B$ in time $t$, $\frac{-dA}{dt} = k_1(A - x)$, where $k_1$ is the fraction of $A$ transferred to $B$ per unit time. Also $\frac{-dB}{dt} = k_2(B + x)$, where $k_2$ is the fraction of $B$ transferred to $A$ per unit time. Now $\frac{-d(A - x)}{dt} = \frac{+dx}{dt} = k_1(A - x) - k_2(B + x)$. Integrating and solving:

$$\ln\left[\frac{k_1A - k_2B}{k_1(A - x) - k_2(B + x)}\right] = (k_1 + k_2)t. \tag{1}$$

At equilibrium $\frac{dx}{dt} = 0$ and $k_1(A - x) = k_2(B + x)$. Then $\frac{k_1}{k_2} = \frac{(B + x)}{(A - x)}$. In our experimental system $(A - x)$ corresponds to the total amount of potassium in the plasma compartment and $(B + x)$ to the total amount of potassium in the cell compartment. If the erythrocytes do not lose or gain total potassium while the radioactive potassium is reaching equilibrium, this becomes:

$$k_1[K^+]_p(1-\text{Hct.}) = k_2[K^+]_c(\text{Hct.}) = \text{a constant, } D, \tag{2}$$

where $k_1$ is that fraction of diluted plasma potassium transferred to the cell compartment per unit time, $[K^+]_p$ the concentration of potassium per liter of diluted plasma, $k_2$ the fraction of cell potassium transferred to the plasma per unit time, and $[K^+]_c$ the concentration of cell potassium per liter of cells.

Considering only the radioactive potassium at any time, $t$, $(A - x)$ corresponds to the diluted plasma radioactive potassium, $K^{42}{}_p$, and $(B + x)$ corresponds to the cell radioactive potassium, $K^{42}{}_c$. Substituting in and manipulating equation (1) gives:

$$\ln\left[\frac{k_1A - k_2B}{D}\right] - \ln\left[\frac{K^{42}{}_p}{[K^+]_p} - \frac{K^{42}{}_c}{[K^+]_c}\right] = (k_1 + k_2)t. \tag{3}$$

The diluted blood potassium specific activity is $\frac{K^{42}{}_b}{[K^+]_b}$, where $K^{42}{}_b$ is the diluted blood radioactive potassium, and $[K^+]_b$ is the concentration of potassium per liter of diluted blood. Changing the logarithms to the base ten:

$$\log\left(\frac{\frac{K^{42}{}_p}{[K^+]_p} - \frac{K^{42}{}_c}{[K^+]_c}}{\frac{K^{42}{}_b}{[K^+]_b}}\right) = \frac{-(k_1 + k_2)t}{2.3} + \log\frac{(k_1A - k_2B)K^{42}{}_b}{D[K^+]_b} \tag{4}$$

Since the last term is a constant, it follows that

$$\log\left(\frac{\frac{K^{42}{}_p}{[K^+]_p} - \frac{K^{42}{}_c}{[K^+]_c}}{\frac{K^{42}{}_b}{[K^+]_b}}\right)$$

plotted against time should be linear. This is true experimentally. The constant slope of this line indicates that the rate of exchange of potassium between the plasma and the cells does not vary throughout the experiment.

Another way of plotting this is log $[\frac{K^{42}_p}{[K^+]_p} - \frac{K^{42}_{p\infty}}{[K^+]_{p\infty}}]$ against time. $\frac{K^{42}_{p\infty}}{[K^+]_{p\infty}}$ is the specific activity of the diluted plasma potassium at equilibrium. This is equal to the specific activity of the diluted blood potassium, $\frac{K^{42}_b}{[K^+]_b}$. As the specific activity of the potassium of the diluted blood and the specific activity of the potassium of the diluted plasma, $\frac{K^{42}_p}{[K^+]_p}$, can be readily determined experimentally, while the specific activity of the potassium of the cells, $\frac{K^{42}_c}{[K^+]_c}$, must be calculated, this method of plotting is more convenient. Fig. 2 shows data for one experiment representative of 18 similar experiments. The slopes of the lines derived from the two methods of plotting are iden-

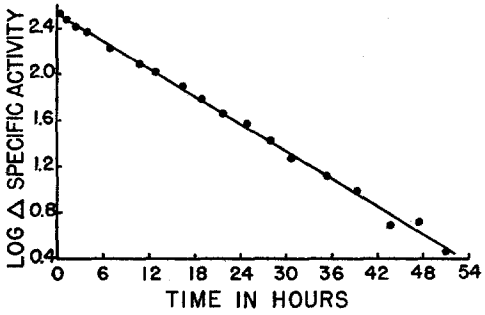


FIG. 2. The log of the difference between the specific activities of the diluted plasma potassium and the diluted blood potassium plotted against time.

tical, but the intercepts are different. These relationships have been pointed out by Sheppard (11).

The slope, $m$, of this line is related to $k_1$ and $k_2$ by the relation:

$$k_1 + k_2 = -2.3m \tag{5}$$

From equations (2) and (5), one may calculate values of $k_1$ and $k_2$.

$$k_2 = \frac{2.3m(1 - \text{Hct.})[K^+]_p}{[K^+]_b} \text{ and } k_1 = -2.3m - k_2.$$

The mean value of $k_2$ thus calculated for 18 experiments in human blood from three donors is 0.0150. Variations between subjects were no greater than for repeated determinations on the same subject.

This experimental system contains diluted whole blood in which both red cells and white cells are present. Sheppard and Martin (12) found in whole canine blood a faster initial exchange rate which was attributed to the frac-

tion containing the white cells. This early fast component was not found in our experiments. The fraction of the whole blood potassium accounted for by the white cells is much lower in human blood than in canine blood. It is felt that the rate calculated above for the exchange of potassium in the human red cell is not appreciably influenced by the presence of white cells.

### *Effect of Varying Temperature*

In each of five experiments the exchange rate was determined at three different temperatures which ranged from 4.8–38.7°C. In contrast to the 34–39° C. temperature range where the incubated cells showed an increase in total intracellular potassium, the cells when incubated at 5°C. show a decrease in intracellular potassium which amounts to about 3 per cent of the initial intracellular potassium in 24 hours. Between 18° and 25°C., there is essentially no gain or loss of cell potassium. Between 18° and 38°C., the calculated intracellular-extracellular potassium gradient was essentially unchanged after initial adjustments had occurred. At 5°C., there is a slow breakdown of the potassium gradient. Data showing these relationships for two experiments are presented in Table III. The loss of potassium from the cells at 2–7°C. has been reported by several workers (10, 13, 14). Its restoration upon warming the cell to 37°C. has been reported by Harris (14).

Exchange rates calculated for the various temperatures are shown in Table IV. The temperature coefficient ($Q_{10}$) of the exchange rate was found to be 2.2 with a standard deviation of 0.2. When the experiments carried out at temperatures ranging from 34–39°C. are corrected to 37°C., the mean exchange rate, $k_2$, is 0.0163. This means that 1.63 per cent of the erythrocyte potassium crosses the cell membrane into the plasma every hour and corresponds to 1.52 millimoles of potassium being transferred in each direction across the red cell membrane per liter of red cells per hour. This is equivalent to about 190 potassium molecules per square micron of erythrocyte area per second. This agrees with the exchange rate obtained by Sheppard and Martin (15). From this exchange constant, the half turnover time, or the time in which half of the potassium molecules leave the red cells and are replaced, can be calculated from the expression $t_{\frac{1}{2}} = \frac{0.693}{k_2}$ to be 43 hours. A summary of the data from these experiments is shown in Table V.

Fig. 3 shows the relationship between the rate of exchange of potassium and the rate of utilization of glucose at three temperatures as determined in one of five experiments. The parallel slopes represent similar temperature coefficients and demonstrate that the ratio of the rate of potassium exchanged to the rate of glucose utilized is the same over the temperature range studied. When the glucose utilization values in our experiments are corrected to 37°C., we find

TABLE III

*Effect of Temperature on the Distribution of Potassium between Diluted Plasma and Red Cells*

| Experiment No. | Temperature | Time | $[K^+]_p$/l. plasma | Hematocrit determination | $[K^+]_p$/l. blood | $[K^+]_c$/l. blood | Change in $[K^+]_c$ | $\frac{[K^+]_c/l.H_2O_c}{[K^+]_p/l.H_2O_p}$ |
|---|---|---|---|---|---|---|---|---|
| | °C. | hrs. | mM/l. | per cent | mM/l. | mM/l. | per cent | |
| 23 | 38.1 | 0.32 | 4.25 | 17.4 | 3.51 | 16.79 | 0 | 33.9 |
| | | 0.85 | 4.21 | 17.8 | 3.46 | 16.84 | +0.3 | 33.2 |
| | | 6.70 | 4.04 | 17.6 | 3.32 | 16.98 | +1.1 | 35.5 |
| | | 12.32 | 4.00 | 18.0 | 3.28 | 17.02 | +1.4 | 34.6 |
| | | 25.08 | 4.00 | 18.6 | 3.26 | 17.04 | +1.5 | 33.0 |
| | 24.4 | 0.38 | 4.41 | 18.3 | 3.60 | 17.20 | 0 | 31.8 |
| | | 1.82 | 4.44 | 19.4 | 3.58 | 17.22 | +0.1 | 28.9 |
| | | 6.77 | 4.42 | 19.2 | 3.58 | 17.22 | +0.1 | 29.4 |
| | | 12.38 | 4.42 | 19.5 | 3.56 | 17.24 | +0.2 | 28.8 |
| | | 25.15 | 4.41 | 19.6 | 3.54 | 17.26 | +0.3 | 28.8 |
| | 4.8 | 0.35 | 4.40 | 19.6 | 3.54 | 17.96 | 0 | 31.2 |
| | | 0.78 | 4.42 | 20.9 | 3.50 | 18.00 | +0.2 | 28.2 |
| | | 6.73 | 4.59 | 20.9 | 3.63 | 17.87 | −0.5 | 27.0 |
| | | 12.35 | 4.89 | 20.6 | 3.88 | 17.62 | −1.9 | 25.5 |
| | | 25.12 | 5.30 | 21.2 | 4.12 | 17.38 | −3.2 | 22.5 |
| 24 | 38.0 | 0.58 | 4.06 | 17.3 | 3.36 | 17.54 | 0 | 37.4 |
| | | 3.90 | 3.93 | 18.6 | 3.20 | 17.70 | +0.9 | 34.9 |
| | | 6.35 | 3.83 | 18.7 | 3.11 | 17.79 | +1.4 | 35.8 |
| | | 12.12 | 3.76 | 18.9 | 3.05 | 17.85 | +1.8 | 36.0 |
| | | 23.15 | 3.67 | 19.7 | 2.94 | 17.96 | +2.4 | 35.0 |
| | 18.0 | 0.40 | 4.09 | 17.7 | 3.37 | 17.23 | 0 | 35.5 |
| | | 3.72 | 4.10 | 19.5 | 3.30 | 17.30 | +0.4 | 30.8 |
| | | 6.17 | 4.15 | 19.3 | 3.35 | 17.25 | +0.1 | 30.8 |
| | | 11.93 | 4.14 | 19.4 | 3.34 | 17.26 | +0.2 | 30.6 |
| | | 22.97 | 4.23 | 19.8 | 3.39 | 17.21 | −0.1 | 29.0 |
| | 5.0 | 0.22 | 4.11 | 18.5 | 3.35 | 17.15 | 0 | 33.7 |
| | | 3.53 | 4.27 | 20.0 | 3.41 | 17.09 | −0.4 | 28.8 |
| | | 5.98 | 4.30 | 20.0 | 3.44 | 17.06 | −0.5 | 28.5 |
| | | 11.75 | 4.55 | 19.8 | 3.65 | 16.85 | −1.8 | 27.0 |
| | | 22.78 | 4.76 | 19.8 | 3.82 | 16.68 | −2.7 | 25.5 |

that about 250 mg. glucose are utilized by a liter of red cells every hour. This is 1.4 millimoles of glucose per hour per liter of red cells.

### *Effect of Varying Conditions*

In the rocker-dilution system we employed, there were some experimental conditions which varied from experiment to experiment. It was not possible

to have an identical percentage of red cells present in all experiments. The potassium concentration of the medium was not exactly the same as the potassium concentration of the donor's plasma. The initial glucose level of the diluted whole blood varied and the glucose concentration was decreasing during the course of any one experiment. The pH was decreasing during incubation. The effects of varying these factors have, therefore, been studied, and the effect of heparin with and without phenol as a preservative has also been investigated.

TABLE IV

*Effect of Temperature on Rate of Potassium Transfer*

| Experiment No. | Temperature | $k_2$ | mM K exchanging per l. RBC per hr. |
|---|---|---|---|
| | °C. | | |
| 23 | 38.1 | 0.0168 | 1.60 |
| | 24.4 | 0.00764 | 0.68 |
| | 4.8 | 0.00167 | 0.14 |
| 24 | 38.0 | 0.0155 | 1.52 |
| | 18.0 | 0.00305 | 0.28 |
| | 5.0 | 0.00135 | 0.18 |
| 25 | 38.5 | 0.0168 | 1.72 |
| | 23.5 | 0.0042 | 0.43 |
| | 7.0 | 0.0031 | 0.31 |
| 26 | 38.7 | 0.0169 | 1.61 |
| | 22.5 | 0.00594 | 0.54 |
| | 7.7 | 0.00292 | 0.26 |
| 27 | 37.7 | 0.0166 | 1.57 |
| | 29.7 | 0.0124 | 1.15 |
| | 22.5 | 0.00556 | 0.51 |

When in two experiments the percentage of red cells was varied from 10 to 45 there was no change in the calculated $k_2$. The potassium concentration of the diluted plasma was varied from 2.02 to 74.4 millimoles per liter. Among six varying levels of potassium in the diluted plasma within this range there was no significant variation from normal in the fraction of erythrocyte potassium exchanging per hour or in the total millimoles of potassium transferred across the membrane per hour. Data from two experiments are shown in Table VI. Varying the initial glucose concentration from 2.5 to 8.0 mg. per ml. also produced no change in the value of $k_2$.

The effect of varying the pH was investigated in three experiments by equilibrating the incubation system with 2.5, 5, 10, and 20 per cent carbon dioxide

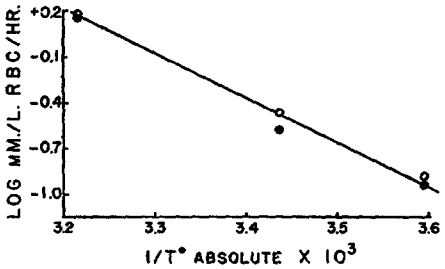


FIG. 3. The log of the millimoles of potassium transferred from a liter of red cells in 1 hour and the log of the millimoles of glucose utilized by a liter of red cells in 1 hour plotted against the reciprocal of the absolute temperature $\times 10^3$. ○ represents millimoles of glucose utilized and ● represents millimoles of RBC potassium exchanged.

TABLE V

*Determination of K Exchange Rate in Human Red Blood Cells*

| Experiment No. | Donor | Temperature | $m$ | $k_2$ | $k_2$ corrected to 37°C. | mM K exchanging per l. RBC per hr. at 37°C |
|---|---|---|---|---|---|---|
| | | °C. | | | | |
| 1 | A | 35.8 | 0.0454 | 0.0156 | 0.0171 | 1.62 |
| 2 | B | 36.0 | 0.0419 | 0.0144 | 0.0156 | 1.46 |
| 3 | A | 36.4 | 0.0356 | 0.0129 | 0.0135 | 1.29 |
| 4 | B | 36.5 | 0.0350 | 0.0128 | 0.0133 | 1.22 |
| 5 | A | 34.4 | 0.0388 | 0.0147 | 0.0180 | 1.61 |
| 6 | A | 34.4 | 0.0388 | 0.0144 | 0.0176 | 1.56 |
| 7 | A | 34.4 | 0.0435 | 0.0155 | 0.0187 | 1.53 |
| 8 | A | 35.0 | 0.0390 | 0.0146 | 0.0171 | 1.56 |
| 9 | A | 35.0 | 0.0390 | 0.0164 | 0.0192 | 1.74 |
| 10 | B | 34.0 | 0.0346 | 0.0137 | 0.0173 | 1.63 |
| 11 | B | 34.0 | 0.0346 | 0.0131 | 0.0165 | 1.55 |
| 12 | A | 38.1 | 0.0430 | 0.0168 | 0.0154 | 1.47 |
| 13 | B | 38.0 | 0.0428 | 0.0155 | 0.0143 | 1.40 |
| 14 | B | 35.0 | 0.0350 | 0.0138 | 0.0161 | 1.54 |
| 15 | C | 35.0 | 0.0397 | 0.0151 | 0.0177 | 1.67 |
| 16 | B | 38.5 | 0.0390 | 0.0168 | 0.0149 | 1.53 |
| 17 | A | 38.7 | 0.0445 | 0.0169 | 0.0148 | 1.41 |
| 18 | A | 37.7 | 0.0440 | 0.0166 | 0.0157 | 1.48 |
| Mean | | | | 0.0150 | 0.0163 | 1.52 |
| Standard deviation | | | | ±0.0014 | ±0.0017 | ±0.12 |

in air. The corresponding initial pH values ranged from 7.5 to 7.7, 7.4 to 7.5, 7.2 to 7.3, and 7.0 to 7.1, respectively. At 24 hours they were 7.2 to 7.4, 7.2 to 7.3, 7.0 to 7.1, and 6.9 to 7.0. Glucose utilization rates were higher at the higher

pH ranges. No significant variations in the exchange rate or in the total millimoles of potassium transferred at these different pH values were found (Table VII). Likewise, no deviations from normal were found when 5 per cent carbon dioxide in air, oxygen, or nitrogen was used for equilibration during incubation.

Because the commerical heparin used as the anticoagulant contained 0.45 per cent phenol as a preservative, three experiments were conducted to compare the effect of heparin containing phenol with heparin without preservative. No significant variations in the exchange rates were found.

TABLE VI

*Effect of Varying Extracellular Potassium Concentration on $k_2$*

| Experiment No. | $[K]^+_p$/l. plasma | $m$ | $k_2$ at 37°C. | mM K exchanging per l. RBC per hr. at 37°C. |
|---|---|---|---|---|
| | mM/l. | | | |
| 19 | 2.02 | 0.0600 | 0.0128 | 1.24 |
| | 3.42 | 0.0411 | 0.0141 | 1.34 |
| | 5.17 | 0.0321 | 0.0147 | 1.42 |
| | 8.02 | 0.0212 | 0.0138 | 1.33 |
| 20 | 4.47 | 0.0397 | 0.0177 | 1.67 |
| | 38.5 | 0.0082 | 0.0140 | 1.33 |
| | 74.4 | 0.0077 | 0.0158 | 1.58 |

TABLE VII

*Effect of Varying pH on $k_2$*

| Experiment No. | $CO_2$ | Initial pH | After 24 hrs. Final pH | $m$ | $k_2$ at 37°C. | mM K exchanging per l. RBC per hr. at 37°C. | Mg. glucose utilized per l. RBC per hr. at 37°C. |
|---|---|---|---|---|---|---|---|
| | *per cent* | | | | | | |
| 21 | 2.5 | 7.5 | 7.4 | 0.0360 | 0.0137 | 1.33 | 243 |
| | 5 | 7.4 | 7.3 | 0.0350 | 0.0133 | 1.27 | 189 |
| | 10 | 7.2 | 7.0 | 0.0380 | 0.0145 | 1.33 | 170 |
| | 20 | 7.0 | 6.9 | 0.0385 | 0.0149 | 1.33 | 159 |
| 22 | 2.5 | 7.7 | 7.2 | 0.0337 | 0.0143 | 1.40 | 200 |
| | 20 | 7.0 | 6.9 | 0.0368 | 0.0159 | 1.43 | 122 |

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## SUMMARY

1. The exchange of potassium by the human erythrocyte has been studied *in vitro* using radioactive potassium.

2. An incubation technique which maintains erythrocytes in an essentially normal state for over 48 hours was employed.

3. Exchange of radioactive potassium between the red cells and the extracellular fluid was regular and progressive, the specific activities of the intra- and extracellular fluids reaching equal values. This indicates that all the erythrocyte potassium is exchangeable and is exchanging at the same rate.

4. From these data, it was calculated that at 37°C., 1.6 per cent of the erythrocyte potassium exchanges per hour, corresponding to an exchange of 1.5 mM of potassium per liter of red cells per hour. The time required for the exchange of 50 per cent of the red cell potassium is calculated to be 43 hours.

5. The temperature coefficient ($Q_{10}$) of the potassium exchange rate is 2.2. This is the same as the temperature coefficient of the rate of utilization of glucose by the human erythrocyte.

6. Varying the percentage of red cells, plasma potassium concentration, initial glucose level, and pH between 7.0 and 7.7 had no effect on the potassium exchange rate.

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