CALCULATION OF THE RATE OF TURNOVER OF A COMPOUND FROM EXCRETION DATA OF A LABELED END-PRODUCT

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The determination of the rate of turnover of a compound in the body by administering a labeled compound as tracer is now a common procedure (1-4). The rate of turnover is usually calculated by plotting the logarithm of the concentration of a labeled end-product excreted in urine against time, but the conditions under which this procedure is justified do not seem to have been fully recognized. Moreover, there are other ways of calculation, the advantages and disadvantages of which, compared with the usual way, do not seem to have been sufficiently appreciated. These ways of calculation and their relative merits will be discussed in this paper.

The rate of turnover of a compound is defined in this paper as the ratio of the amount \( R \) of the compound removed (from the metabolic pool) and replaced per unit time to the total amount \( P \) of the compound in the pool. Thus defined, the rate of turnover \( K = \frac{R}{P} \) is a rate constant in a first-order reaction.

It can be shown\(^1\) that in a series of consecutive first-order reactions (or processes), the cumulative amount \( e \) of an end-product at time \( t \) is given by the equation:

\[
e = \alpha e^{-\alpha t} \left[ 1 - \left( \alpha e^{-\alpha t} + \beta e^{-\beta t} + \cdots + s e^{-st} \right) \right]
\]

\( ^1 \) The kinetics of consecutive, first order, irreversible reactions has been worked out by Thiersch (Thiersch, F., Z. physik. Chem., 1924, 111, 175). However, equation (1) in the present paper differs from that given by Thiersch (Equation 14 in his paper) in the presence of the term \( \alpha e^{-\alpha t} \). In Thiersch's formulation, each intermediate compound is converted into a single compound in the next step. In a biochemical process an intermediate compound may be changed in several ways (physical or chemical) in parallel reactions, and only a fraction of the total change is due to the process which leads to the end-product under consideration. A constant factor, which may be called the fraction, therefore enters into the rate equation for each step. In equation (1) \( \alpha e^{-\alpha t} \) is the product of all the fractions in the preceding steps. A fuller treatment of the kinetics of consecutive reactions in biochemical processes will be reported in another paper.
RATE OF TURNOVER OF COMPOUND FROM EXCRETION DATA

where \( A_0 \) = amount of the original labeled compound administered, e.g., in the experiments reported in the following paper, \( A_0 \) is the amount of aspartic acid-N\(^{15} \) fed.

\( K_A, K_B, \ldots \) rate constants in the consecutive reactions or processes from the absorption of the labeled aspartic acid to the excretion of urea-N\(^{15} \) in urine.

\( \alpha_B = \) the fraction of \( A_0 \) which appears as the end-product, in the present example as urea-N\(^{15} \).

\( a, b, c, \ldots \) are constants.

When one of the rate constants, \( K_n \), is smaller than all the others, there will come a time when \( e^{-K_st} \) alone in equation (1) is significant while all the other exponential terms are negligible. Equation (1) then reduces to

\[
e = \alpha_B A_0 (1 - se^{-K_st})
\]

Differentiating equation (2), we obtain

\[
\frac{de}{dt} = \alpha_B A_0 K_s e^{-K_st}
\]

But

\[
\frac{de}{dt} = \tau E
\]

Where \( \tau \) is the "isotopic ratio", that is, \( \frac{N^{15}}{N} \) of urea-N in the above example. The isotopic ratio in the pool is assumed to be the same as that in urine.

\( E \) is the amount of the end-product, in the above example, urea-N excreted per unit time.

Therefore,

\[
\tau = \frac{\alpha_B A_0 K_s}{E} e^{-K_st}
\]

and

\[
se^{-K_st} = \frac{\tau E}{\alpha_B A_0 K_s}
\]

Substituting equation (5 a) in (2) and rearranging, we obtain

\[
\tau = \frac{\alpha_B A_0 K_s}{E} e^{-K_st} - \frac{\tau}{E}
\]

and

\[
\alpha_B = \frac{E}{A_0} \left( \frac{\epsilon}{\tau} + \frac{\tau}{K_s} \right)
\]

These equations are the basis of all the methods of calculation described below.
The value of $K_r$ calculated on the basis of the above equations is the smallest rate constant in a series of consecutive chemical reactions and physical processes (diffusion) which precede the appearance of the end-product in urine. The smallest rate constant may well be the rate of turnover of the compound in question, but it cannot be overemphasized that this is an assumption which must be justified by previous knowledge of the relative magnitudes of the rate constants involved in the consecutive reactions and processes.$^3$

The methods of calculation are summarized in Table I. The methods differ depending on whether $e$ or $r$ data are used (that is, whether $K$ enters into the calculations or not) and on whether non-cumulative ($r, \Delta r, \Delta e$) or cumulative ($\sum (r\Delta t), e$) values are used.

I. Semilogarithmic plot—with non-cumulative values.

From equation (5) it is seen that if $\ln r$ is plotted against $t$, the result is a straight line, the slope of which is $-K_r$.$^3$

In plotting this curve, it is a common practice to use the average $r$ ($\bar{r}$) of an interval as the instantaneous $r$ at the middle point of the interval. This practice is justified only when the intervals are short and $K_r$ is numerically small.

$^3$ For example, after feeding creatine labeled with N$^{15}$, the rate of turnover of creatine was calculated from the rate of fall of N$^{15}$ concentration of creatinine in urine (1). Assuming that all the creatine is in the muscle, there are at least three steps between creatine in muscle and creatinine in urine, namely:

- Creatine in muscle $\rightarrow$ creatinine in muscle
- Creatine in muscle $\rightarrow$ creatinine in blood
- Creatinine in blood $\rightarrow$ creatinine in urine

The rate constant of 0.0164 day$^{-1}$ obtained by Hoberman, Sims, and Peters may be the rate constant for any one of the three steps. From the excretion data of creatinine alone in the experiment mentioned, it is impossible to make a decision. However, the decisions can be made on the basis of other knowledge. From the fact that creatinine in blood is of the order of 1.5 mg. per 100 ml. blood and the amount of creatinine excreted per day is of the order of 1.6 gm., and assuming 6 liters to be the volume of blood, the rate constant for the third step is at least

$$\frac{1600}{6 \times 15} = 18 \text{ day}^{-1}$$

If the permeability of the muscle to creatinine is of the same order as that of the kidney, the rate constant for the second step must be also of the order of 18 or, in any case, much larger than 0.0164. The conclusion that this value is the rate constant of the first step, namely, creatine $\rightarrow$ creatinine, is therefore correct, but the conclusion is made possible only on the basis of other knowledge regarding the relative magnitudes of the rate constants of the consecutive processes involved.

$^3$ In calculating the slope of the curve plotted with log instead of $\ln$, log is, of course, multiplied by 2.3 to obtain $\ln$. 

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[Note: The paper seems to be discussing a scientific method for calculating rate constants, possibly in the field of biochemistry, and provides a detailed explanation of how to calculate the rate constant $K_r$ based on a series of consecutive chemical reactions and physical processes.]
<table>
<thead>
<tr>
<th>Method</th>
<th>Data</th>
<th>Equation</th>
<th>Example of calculation*</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>I. Semilogarithmic plot, non-cumulative</td>
<td>log ( r ) vs. ( t )</td>
<td>( \ln r = \ln \left( \frac{e^{A_{0}}}{E} \right) - K \cdot t )</td>
<td>I(( e )) 0.0426†</td>
<td>Points fall on two lines (Fig. 1).</td>
</tr>
<tr>
<td></td>
<td>( \log (\Delta e) ) vs. ( t )</td>
<td>( \ln (\Delta e) = \ln \left( \frac{e^{A_{0}}}{E} \right) - K \cdot t )</td>
<td>I(( e )) 0.0367†</td>
<td></td>
</tr>
<tr>
<td>II. Semilogarithmic plot, cumulative</td>
<td>( \log \left[ \frac{e^{A_{0}}}{E} - \Sigma (\Delta t) \right] ) vs. ( t )</td>
<td>( \ln \left[ \frac{e^{A_{0}}}{E} - \Sigma (\Delta t) \right] = \ln \left( \frac{e^{A_{0}}}{E} \right) - K \cdot t )</td>
<td>II(( e )) 0.0363</td>
<td>Points fall on one line (Fig. 2).</td>
</tr>
<tr>
<td></td>
<td>( \log \left( 1 - \frac{e}{e^{A_{0}}} \right) ) vs. ( t )</td>
<td>( \ln \left( 1 - \frac{e}{e^{A_{0}}} \right) = \ln s - K \cdot t )</td>
<td>II(( e )) 0.0435</td>
<td></td>
</tr>
<tr>
<td>III. Arithmetic plot</td>
<td>( r ) vs. ( \Sigma (\Delta t) )</td>
<td>( r = \frac{e^{A_{0}}}{E} - K \cdot \Sigma (\Delta t) ) K</td>
<td>III(( e )) 0.0434‡</td>
<td>Points fall on two lines (Figs. 3–4).</td>
</tr>
<tr>
<td></td>
<td>( r ) vs. ( e )</td>
<td>( r = \frac{e^{A_{0}}}{E} - K \cdot \frac{e}{E} )</td>
<td>III(( e )) 0.0435‡</td>
<td></td>
</tr>
</tbody>
</table>

* Using excretion data of urea-\( 14 \)N of subject R in an experiment reported in the following paper.
† Weighted average.
in relation to the time interval. It can be shown, however, that by using \( r \) of equal consecutive intervals, the error of using \( r \) for \( r \) is reduced to the minimum (Appendix I). With unequal intervals, the error may be considerable.

When urine has been collected at short regular intervals, the \( r \) values obtained experimentally can be plotted directly. When the urine is collected at irregular intervals, \( r \) for equal intervals can be obtained by the following procedure:

1. Calculate the value of \( r \Delta t \) for each interval from the beginning (Table II).
2. Add the \( r \Delta t \) terms to obtain \( \Sigma (r \Delta t) \) up to time at the end of each interval.
3. Plot \( \Sigma (r \Delta t) \) against \( t \) and draw a smooth curve.
4. On the \( \Sigma (r \Delta t) \) curve read \( \Sigma (r \Delta t) \) values for consecutive points placed at equal intervals (say, 3 hours)\(^4\) (Table III).
5. Calculate \( r \Delta t \) for each interval by difference.
6. Divide \( r \Delta t \) thus obtained by \( \Delta t \) to obtain \( r \) for equal intervals.
7. Plot \( r \) against middle point of interval and draw a smooth curve.

Since \( r \) is usually expressed as atom per cent excess, it is convenient to multiply everything by 100 in the above calculations.

\(^4\) Theoretically, the shorter the interval, the more closely does \( r \) approximate \( r \). But from the practical standpoint, the error in \( r \) obtained from the \( \Sigma (r \Delta t) \) curve is greater, the shorter the interval. In the experiment cited for illustration, intervals of 3 to 6 hours are most suitable.

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**TABLE II**

Original Data of Urea-N\(^{15}\) Excretion for Unequal Intervals

<table>
<thead>
<tr>
<th>Time at end of interval (hr.)</th>
<th>( 100 \ r )</th>
<th>( \Delta ) Urea-N(^{15}) excreted in interval (gm.)</th>
<th>( \frac{\Delta \text{Rate of Urea-N}(^{15}) excretion}}{\Delta t} ) (gm./hr.)</th>
<th>( \Delta ) Urea-N(^{15}) excreted up to end of interval (mg.*)</th>
<th>( 100 \ r \Delta t )</th>
<th>( \Sigma (100 \ r \Delta t) )</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.83</td>
<td>0.033</td>
<td>0.278</td>
<td>0.334</td>
<td>0.092</td>
<td>0.092</td>
<td>0.027</td>
</tr>
<tr>
<td>2.00</td>
<td>0.354</td>
<td>0.302</td>
<td>0.538</td>
<td>0.630</td>
<td>0.178</td>
<td>0.205</td>
</tr>
<tr>
<td>3.38</td>
<td>0.168</td>
<td>0.418</td>
<td>0.303</td>
<td>1.333</td>
<td>0.232</td>
<td>0.437</td>
</tr>
<tr>
<td>5.00</td>
<td>0.170</td>
<td>0.657</td>
<td>0.386</td>
<td>2.453</td>
<td>0.289</td>
<td>0.726</td>
</tr>
<tr>
<td>7.00</td>
<td>0.150</td>
<td>0.618</td>
<td>0.322</td>
<td>3.378</td>
<td>0.288</td>
<td>1.014</td>
</tr>
<tr>
<td>12.08</td>
<td>0.124</td>
<td>1.753</td>
<td>0.345</td>
<td>5.558</td>
<td>0.630</td>
<td>1.644</td>
</tr>
<tr>
<td>21.25</td>
<td>0.092</td>
<td>3.358</td>
<td>0.373</td>
<td>8.658</td>
<td>0.845</td>
<td>2.489</td>
</tr>
<tr>
<td>25.50</td>
<td>0.074</td>
<td>1.757</td>
<td>0.417</td>
<td>9.548</td>
<td>0.314</td>
<td>2.803</td>
</tr>
<tr>
<td>30.42</td>
<td>0.059</td>
<td>1.899</td>
<td>0.386</td>
<td>1.290</td>
<td>0.290</td>
<td>3.093</td>
</tr>
<tr>
<td>36.75</td>
<td>0.037</td>
<td>2.544</td>
<td>0.450</td>
<td>1.650</td>
<td>0.234</td>
<td>3.327</td>
</tr>
</tbody>
</table>

* As \( \text{N}\(^{14}\).
Since the amount of the end-product excreted in an interval $\Delta t$ is

$$\Delta e = i \Delta t e$$

and $i$ is equivalent to $r$ at the middle point of the interval (Appendix I)

$$\therefore \Delta e = \alpha e A0 K_0 \Delta t e^{-K_0 t}$$

where $t$ is the time at the middle of the interval. Therefore, when $\ln(\Delta e)$ is plotted against $t$, the result is a straight line the slope of which is $-K_0$. The value of $\Delta e$ for equal intervals can be obtained from a $e$ curve in the same manner as $i \Delta t$ is obtained from the $\Sigma(\epsilon \Delta t)$ curve.

### TABLE III

**Derived Data of Urea-$N_{15}$ Excretion for Equal Intervals**

<table>
<thead>
<tr>
<th>Time</th>
<th>mg. N$^{15}$</th>
<th>$\Delta e$ by difference</th>
<th>$\Sigma(100 e \Delta t)$ by difference</th>
<th>$\Delta e$ from $\Sigma e$ curve</th>
<th>$100 \epsilon$ from $100 e \Delta t$ curve</th>
</tr>
</thead>
<tbody>
<tr>
<td>hrs.</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>1.25</td>
<td>1.68</td>
<td>0.38</td>
<td>0.48</td>
<td>1.54</td>
</tr>
<tr>
<td>6</td>
<td>2.93</td>
<td>1.37</td>
<td>0.86</td>
<td>0.41</td>
<td>1.32</td>
</tr>
<tr>
<td>9</td>
<td>4.30</td>
<td>1.25</td>
<td>1.27</td>
<td>0.35</td>
<td>1.18</td>
</tr>
<tr>
<td>12</td>
<td>5.55</td>
<td>1.12</td>
<td>1.62</td>
<td>0.31</td>
<td>1.07</td>
</tr>
<tr>
<td>15</td>
<td>6.67</td>
<td>1.03</td>
<td>1.93</td>
<td>0.28</td>
<td>0.96</td>
</tr>
<tr>
<td>18</td>
<td>7.70</td>
<td>0.90</td>
<td>2.21</td>
<td>0.25</td>
<td>0.96</td>
</tr>
<tr>
<td>21</td>
<td>8.60</td>
<td>0.85</td>
<td>2.46</td>
<td>0.22</td>
<td>0.86</td>
</tr>
<tr>
<td>24</td>
<td>9.45</td>
<td>0.75</td>
<td>2.68</td>
<td>0.20</td>
<td>0.78</td>
</tr>
<tr>
<td>27</td>
<td>10.20</td>
<td>0.65</td>
<td>2.88</td>
<td>0.17</td>
<td>0.70</td>
</tr>
<tr>
<td>30</td>
<td>10.85</td>
<td>0.65</td>
<td>3.05</td>
<td>0.17</td>
<td>0.63</td>
</tr>
</tbody>
</table>

**II. Semilogarithmic plot**—with cumulative values.

From equation (2) it is seen that when $\ln \left(1 - \frac{e}{\alpha e A0}\right)$ is plotted against $t$, the result is a straight line if the correct value of $\alpha e$ is chosen. If $\frac{e}{E}$ is used instead of $e$, for the reason to be mentioned below, $\ln \left(\frac{\alpha e A0}{E} - \frac{e}{E}\right)$ similarly plotted will also give a straight line. In either case, the slope of the curve is $-K_0$. 
III. Arithmetic plot. From equation (6) it is seen that if \( r \) is plotted as ordinate against \( \frac{s}{E} = \Sigma (\bar{r} \Delta t) \) as abscissa the result is a straight line, the slope of which is \(- K_s\) and the intercept is \( \frac{\alpha \Sigma A_0}{E} K_s \), from which the value of \( \alpha_s \) can be calculated.

As an alternative, \( r \) may be plotted against \( e \), the result is a straight line, the slope of which is \(- \frac{K_e}{E} \) and the intercept is \( \frac{\alpha \Sigma A_0}{E} K_s \).

Choice between \( r \) and \( e \) Data

Although \( K_s \) can be evaluated either from \( r \) or \( e \) data, the former is more directly related to \( K_e \) than the latter. The rate of fall of \( r \) is a direct measure of \( K_s \), but the rate of fall of \( \Delta e \) is a measure of \( K_e \) only when \( E \) remains constant. If over a period of time \( E \) is increasing (e.g. see Table II) the value of \( K_s \) evaluated from \( \Delta e \) data will be lower than that evaluated from \( r \) data. On general principles, \( r \) should give more reliable results. Moreover, it is generally observed that \( r \) values are more consistent than \( \Delta e \) values. This can be shown by plotting \( \Delta e \) and \( r \) against the time at the middle point of the interval. The \( r \) points usually fall on a smooth curve while the \( \Delta e \) points are usually more scattered. The lesser consistency of \( \Delta e \) data is due to the inconstancy of \( E \) which enters into the calculation of \( \Delta e \). For this reason, \( r \) data are preferable to \( e \) data for the evaluation of \( K_s \).

In Table I, the values of \( K_s \) calculated from \( e \) data are lower than those calculated from \( r \) data, because from 6 to 30 hours the general trend of \( E \) was increasing which made \( \Delta e \) fall more slowly than \( r \).

Although \( K_s \) can be evaluated without using \( e \) data, the latter are required to calculate \( \alpha_s \). Since \( E \) is subject to some variation, it is better to use \( \frac{\bar{e}}{E} \), the amount of the labeled end-product excreted per unit rate of total excretion of the end-product, instead of \( e \). Since \( \Delta e = \bar{r} \Delta t \), therefore

\[
\frac{\bar{e}}{E} = \frac{\Sigma (\Delta e)}{\Sigma (\bar{r} \Delta t)}
\]

The last value can be calculated without reference to \( E \) in different intervals. In calculating \( \alpha_s \) from \( \frac{\alpha \Sigma A_0}{E} \), the average \( E \) is used.

Choices between Non-Cumulative and Cumulative Values

Deviations are more easily seen with non-cumulative values—\( r, \bar{r}, \bar{r} \Delta t, \) and \( \Delta e \), than with cumulative values, \( \Sigma (\bar{r} \Delta t) \) and \( e \). When \( \Delta e \) or \( \bar{r} \) is plotted against time, an obvious error can be recognized and the value rejected. When \( e \) or
\[ \Sigma(\Delta t) \] is plotted against time, even relatively poor data may give a fairly smooth curve. For this reason, \( \Delta \varepsilon \) or \( \Sigma(\Delta t) \) obtained by difference should be tested for consistency by plotting against time, even though the \( \varepsilon \) or \( \Sigma(\Delta t) \) curve appears to be smooth.

With non-cumulative values, an error in one point has no influence on other points. With cumulative values an error in one point remains with all subsequent points. It is preferable, therefore, to use non-cumulative values for the evaluation of rate constants.

![Graph](image)

**Fig. 1. Evaluation of \( K \), by method I.** Upper curve, log (\( \Delta \varepsilon \)) for 3 hour intervals vs. \( t \) at middle point. Lower curve, log (100 \( \tau \)) vs. \( t \). The point 1 on the ordinate corresponds to \( \Delta \varepsilon = 1 \) mg, \( N^{10} \) for the log (\( \Delta \varepsilon \)) curve, and to 100 \( \tau = 0.100 \) atom percent excess for the log (100 \( \tau \)) curve.

The slope of the log (\( \Delta \varepsilon \)) curve between 6 to 9 hours is 0.0541; between 9 to 30 hours, 0.0337. Weighted average for the period 6 to 30 hours, 0.0363.

The slope of the log (100 \( \tau \)) curve between 6 to 12 hours is 0.0491; between 12 to 30 hours, 0.0404. Weighted average for the period 6 to 30 hours, 0.0426.

**Choice among Methods I, II, and III**

Since all the above methods of calculation are based on the same basic equation, they should give the same result when experimental data fit the theory perfectly. When this is not the case, the value of \( K \), obtained is an average which will differ somewhat with the method used.

Method III gives the most information because it is the most sensitive. Deviations of \( K \) from constancy can be easily seen on the arithmetic plot.
Method I is a little less sensitive, because logarithmic plot tends to minimize the appearance of deviations. Method II is the least sensitive, because the appearance of deviation is further minimized by the use of cumulative values (Figs. 1–4).

In the example taken for illustration, the points in the curve \( r \) against \( \frac{e}{E} \) fall on two straight lines (Fig. 3). The point of change of \( K_s \) occurred about 12 hours after feeding. A similar picture is given by the curve \( \log r \) against \( t \) (Fig. 1). All but one point (at 6 hours) in the curve \( \log r \) against \( t \) fall on a straight line. The curves obtained with method II (Fig. 2) show no change of \( K_s \). The points all fall closely on a straight line, but this is because the method...
Fig. 3. Evaluation of \( K_s \) by plotting \( r \) against \( \Sigma (r \Delta t) \). Slope of curve between 6 to 15 hours, 0.0498; between 15 to 30 hours, 0.0395. Weighted average, 0.0434. The first point is at \( t = 6 \). Subsequent points are spaced 3 hours apart.

Fig. 4. Evaluation of \( K_s \) by plotting \( r \) against \( e \). Slope of curve, \(-\frac{K_s}{E}\), between 6 to 12 hours is \( \frac{0.01457}{100} \). \( E = 341 \text{ mg. per hour. } K_s = 0.0497 \). Slope of curve between 12 to 30 hours, \( \frac{0.01086}{100} \). \( E = 381 \text{ mg. per hour. } K_s = 0.0414 \). Weighted average \( K_s \) for the period 6 to 30 hours is 0.0435. The first point is at \( t = 6 \). Subsequent points are spaced 3 hours apart.
is not sensitive. If the value of $\alpha_s$ assumed is within 10 per cent of the correct value, any curve would appear straight enough to the eye. Without special care, the correct answer may be easily missed. Method II is attractive as a way of obtaining an average $K_s$ when it does not remain constant, but it has the disadvantage that it is a "by-trial" method. Several trials may be necessary before the final answer is obtained. With the other methods, the answer is obtained in one operation. Method III has the advantage that it gives both $K_s$ and $\alpha_s$ in one plotting, while with the other methods, $K_s$ is first evaluated and $\alpha_s$ is then calculated with equation (6 a).

Instead of using graphic methods, $K_s$ may be evaluated by algebraic methods. For instance, instead of method I, $K_s$ may be calculated from the ratios of $r$ for consecutive points or those of $\bar{r}$ or $\Delta r$ for consecutive intervals ($\Delta t$). For it can be shown that for $r_1, r_2, r_3 \ldots \ldots \text{at} \ t_1, t_2, t_3 \ldots \ldots$

$$\frac{r_1}{r_2} = \frac{r_3}{r_1} = \ldots \ldots = e^{K_s \Delta t} \tag{7}$$

and similar relations hold for $\Delta r$ and $\bar{r}$. Instead of method III, $K_s$ can be calculated from the relation

$$\frac{r_1 - r_2}{r_2 \Delta t} = K_s \tag{8}$$

which can be derived from equation (5).

With equation (7) or (8) $K_s$ can be calculated for each interval and the results averaged. Deviations of $K_s$ from constancy are more easily seen with these algebraic methods than with the graphic methods. But they are too sensitive; errors in $r$ become unduly important. While the algebraic methods are capable of high precision when the data are precise enough to justify their use, they have no advantage over the graphic methods with data of ordinary precision.

SUMMARY

Methods of calculation of the rate of turnover of a compound from excretion data of a labeled end-product are compared and their relative merits discussed. In this calculation, it is assumed that the rate of turnover of the compound in question is the smallest rate constant in a series of consecutive reactions or processes.

BIBLIOGRAPHY


* For those experiments reported in the following paper, in which method III shows two values of $K_s$, method II would give a single value.
Justification for the Use of the Average \( r \) of an Interval as the Instantaneous \( r \) at the Middle Point of the Interval

From equation (2) it follows that

\[
e_1 = \alpha A_0 (1 - e^{-\mathcal{E}_A s t_1}) \\
e_2 = \alpha A_0 (1 - e^{-\mathcal{E}_A s t_2})
\]

and

\[
\Delta e_{1-2} = e_2 - e_1 = \alpha A_0 e_0 (e^{-\mathcal{E}_A s t_1} - e^{-\mathcal{E}_A s t_2})
\]

but

\[
\Delta e_{1-2} = \frac{\alpha A_0 e_0}{E} (e^{-\mathcal{E}_A s t_1} - e^{-\mathcal{E}_A s t_2})
\]

From equation (5) it follows that the \( r \) at the middle point \( \frac{t_1 + t_2}{2} \) of an interval \( t_1 \) to \( t_2 \) is

\[
r \left( \frac{t_1 + t_2}{2} \right) = \frac{\alpha A_0 e_0 e^{-\mathcal{E}_A s \left( \frac{t_1 + t_2}{2} \right)}}{E} \]

Dividing equation (10) by equation (11), we obtain

\[
\frac{\frac{\Delta e_{1-2}}{e^{-\mathcal{E}_A s t_2}}}{\frac{\Delta e_{1-2}}{e^{-\mathcal{E}_A s t_2}}} = \frac{\frac{\alpha A_0 e_0}{E} (e^{-\mathcal{E}_A s t_1} - e^{-\mathcal{E}_A s t_2})}{\frac{\alpha A_0 e_0}{E} (e^{-\mathcal{E}_A s t_1} - e^{-\mathcal{E}_A s t_2})}
\]

By expanding \( e^{-\mathcal{E}_A s t_1} \), \( e^{-\mathcal{E}_A s t_2} \), and \( e^{-\mathcal{E}_A s (t_1 + t_2)/2} \) each into a series, it can be shown that

\[
\frac{r_{t_1-t_2}}{r_{(t_1+t_2)/2}} = 1 + \frac{K_e (t_2 - t_1)^2}{24} \text{ approximately.}
\]

When both \( K_e \) and \( t_2 - t_1 \) are small, \( K_e (t_2 - t_1)^2 \) is negligible in comparison with 1, and \( r_{t_1-t_2} = r_{(t_1+t_2)/2} \).

When \( K_e = 0.05 \), \( t_2 - t_1 = 3, 6, 9, \) and 12, \( e^{-\mathcal{E}_A s t_1} - e^{-\mathcal{E}_A s t_2} \) is larger than \( K_e e^{-\mathcal{E}_A s (t_1+t_2)/2} \) by 0.1, 0.4, 0.9, and 1.5 per cent respectively. When \( K_e = 0.10 \), the corresponding differences are 0.4, 1.5, 3.3, and 5.8 per cent respectively.
Thus, when \( K_s \) is less than 0.1 and the interval \( t_2 - t_1 \) is less than 6, or when the product of \( K_s \) and \( (t_2 - t_1) \) is less than 0.6, the average \( r \) of an interval may be taken as the instantaneous \( r \) at the middle point of the interval without appreciable error.

From equation (13) it is seen that for a series of equal intervals, the percentage error incurred in using \( \tilde{r} \) for \( r \) is the same for all intervals. Thus, when \( \ln \tilde{r} \) is used instead of \( \ln r \) in method I, there is a constant error of

\[
\ln \left[ 1 + \frac{K_s(t_2 - t_1)^2}{24} \right].
\]

Even when this error is appreciable, only the intercept will be in error, while the slope will still be correct. If the intervals are not equal, the slope will also be in error.