

## APPENDIX

## Calculation of Rate of Exchange Transamination

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The problem was to determine the rate of exchange transamination between glutamate and  $\alpha$ -oxoglutarate when the situation was complicated by a net conversion of glutamate into  $\alpha$ -oxoglutarate and by the continuous utilization of  $\alpha$ -oxoglutarate.

The diagram shown in Scheme 1 is a simplification of what occurred when the above substrates were incubated with tissue preparations. The following simplifications were made:

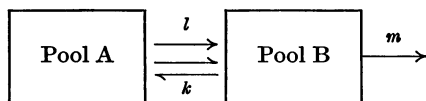
(i) Net glutamate removal only occurred via its conversion into  $\alpha$ -oxoglutarate.

(ii) Pools A and B were homogeneous.

(iii) The rates  $k$ ,  $l$  and  $m$  were constant and independent of the amounts of substrate within the ranges studied.

Rate of change in size of pool A:

$$\frac{da}{dt} = -l \quad (1)$$



|                        | Pool A<br>(glutamate) | Pool B<br>( $\alpha$ -oxoglutarate) |
|------------------------|-----------------------|-------------------------------------|
| Amount of substrate... | ... $a$               | ... $b$                             |
| Radioactivity...       | ... $x$               | ... $y$                             |
| Specific activity ...  | ... $x/a$             | ... $y/b$                           |

Scheme 1.  $k$ , Rate of exchange transamination;  $l$ , net rate of removal of glutamate;  $m$ , rate of removal of  $\alpha$ -oxoglutarate, calculated as the net decrease in glutamate +  $\alpha$ -oxoglutarate.

Rate of change in size of pool B:

$$\frac{db}{dt} = l - m \quad (2)$$

Rate of change in radioactivity of pool A:

$$\frac{dx}{dt} = \frac{ky}{b} - (k+l)\frac{x}{a} \quad (3)$$

Rate of change in radioactivity of pool B:

$$\frac{dy}{dt} = (k+l)\frac{x}{a} - (k+m)\frac{y}{b} \quad (4)$$

From eqns. (1) and (2):

$$l = \frac{a_0 - a}{t}$$

$$m = l + \frac{b_0 - b}{t}$$

The effect of experimental error was minimized by estimating the values of  $a$  and  $b$  from regression lines; hence  $l$  and  $m$  were determined (Table 3 of Balázs & Haslam, 1965).

An Elliott 803 computer was used to integrate the four simultaneous differential equations and to give values for  $x$  and  $y$  at time  $t$ . The Runge-Kutta-Merson method of integration was employed (Lance, 1960) and the integration was repeated with various values of  $k$ .

The specific activity of each pool was plotted against time for both the experimental results and for the values calculated from the various estimates of  $k$  (Fig. 1 of Balázs & Haslam, 1965). These graphs enable a best estimate of  $k$  to be made.

## REFERENCES

- Balázs, R. & Haslam, R. J. (1965). *Biochem. J.* **94**, 131.  
Lance, G. N. (1960). *Numerical Methods for High Speed Computers*, p. 56. London: Iliffe and Sons Ltd.

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