Further Study on the Turnover Calculation of Lung Lecithins

KIMIYOSHI OHNO, TOYOAKI AKINO and HIDEO KANOH

The First Division, Department of Biochemistry, Sapporo Medical College, Sapporo


Using the data reported previously by Moriya and Kanoh, the metabolic turnover of rat lung lecithins was calculated on the basis of the new assumption that the formation rate ($V_a$) of lecithins from diacylglycerols did not equal the disappearance rate ($V_b$) of lecithins because of the presence of precursors other than diacylglycerols. The ratio of $V_a/V_b$ was almost the same in dienoic lecithin as that for total lecithins, whereas a very low ratio was found in saturated and hexaenoic species. The turnover time of lung lecithins estimated was markedly shorter than those determined in much later experimental periods after administration of labeled precursors. The reason for this discrepancy was discussed.

turnover time; turnover rate; lung lecithin

In vivo studies with different labeled precursors such as glycerol, palmitate and choline have been made to elucidate the metabolic turnover of lung tissue and alveolar lecithins, namely, the physiological metabolism of surfactant (dipalmitoyllecithin) (Harlan et al. 1964; Thomas and Rhodes 1970; Tierney et al. 1967; Spitzer and Norman 1971; Young and Tierney 1972).

We have estimated the turnover rate of lung lecithins in the early experimental periods after administration of labeled precursors (Moriya and Kanoh 1974) and that of lung tissue and alveolar lecithins in the later experimental periods (Toshima et al. 1972). However, the calculation of turnover of lung lecithins in the early experimental periods was carried out by assuming that the rate of formation of lecithins from diacylglycerols equals the rate of disappearance of lecithins. Since recent findings strongly suggest the presence of precursors other than diacylglycerols in the formation of lung lecithins (Akino et al. 1971; Frosolono et al. 1971; Vereyken et al. 1972; Hallman and Raivio 1974; Hasegawa-Sasaki and Ohno 1975), the turnover rates of lung lecithins estimated in the previous experiments have to be corrected by the new assumption. This communication, therefore, focused upon the further calculation of turnover rate of lung lecithins using the data reported before (Moriya and Kanoh 1974).

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Calculation Methods

The metabolic turnover of lecithin species was calculated from the data of in vivo experiment by Moriya and Kanoh (1974), according to the equation of Zilversmit, Entenman and Fishier (1943) and of Wise and Elwyn (1965).

In the reaction scheme illustrated in Fig. 1, it was assumed that the rate of formation of lecithins from diacylglycerols ($V_a$, $\mu$moles·min$^{-1}$·g$^{-1}$ wet tissue) did not equal the rate of disappearance or turnover rate of lecithin ($V_b$, $\mu$moles·min$^{-1}$·g$^{-1}$) because of the possible presence of precursors other than diacylglycerols leading to the formation of lecithins. It was also assumed that the influx of radioactivity into molecular species of lecithins was solely from the corresponding species of diacylglycerols during the periods of 2–30 min, and the contributions of other precursors could be neglected since the specific radioactivities of other precursors such as fatty acids, lysolecithins and phosphatidylethanolamines were much lower than that of diacylglycerol species. However, this assumption was not valid in the case of tetraenoic species, because of the very low specific radioactivity of tetraenoic diacylglycerols and a very rapid equilibration between tetraenoic lecithin and diacylglycerol as shown in the previous paper (Moriya and Kanoh 1974).

Radioactive precursor(s) $\rightarrow$ Diacylglycerols $\rightarrow$ Lecithins $\rightarrow$

$V_a$  $\rightarrow$ $V_b$

$A$  $\rightarrow$ $B$

$\uparrow V_{ai}$

Other precursor(s)

$V_a + \sum_{i=1}^{n} V_{ai} = V_b$: turnover rate of lecithins

$A_i$ (i=1, 2, ..., n)

$\frac{V_a}{V_b} < 1$

Fig. 1. Reaction scheme for the calculation of turnover rate of lecithins.

The basic equation used is

$$\frac{dS_B}{dt} = \frac{V_a \cdot S_A + \sum_{i=1}^{n} V_{ai} \cdot S_{Ai} - V_b \cdot S_B}{B}$$

(1)

where $S_A$, $S_{Ai}$, and $S_B$ are the specific radioactivities (dpm $\mu$ mole$^{-1}$) of diacylglycerols, other precursors and lecithins, respectively, and B is the pool size of rat lung lecithins ($\mu$moles·g$^{-1}$ wet tissue). The following equation is derived from Eq. 1 because of $S_{Ai} \ll S_A$.

$$\frac{dS_B}{dt} = \frac{V_a \cdot S_A - V_b \cdot S_B}{B}$$

(2)

From Eq. 2, we get:
where \( t \) is the experimental time period (min) and \( T_t \) is the turnover time (min).

Eq. 3 can be transformed to give the ratio of the rate of formation of lecithins from diacylglycerols \( (V_a) \) and the turnover rate of lecithin \( (V_b) \) as follows:

\[
\frac{V_a}{V_b} = \int_{t_1}^{t_2} dS_B \cdot \frac{\int_{t_1}^{t_2} S_A \cdot dt - \int_{t_1}^{t_2} S_B \cdot dt}{\int_{t_1}^{t_2} dS_B}
\]

The turnover time of lecithin species \( (T_t) \) can be calculated from Eq. 3, using the ratio \( (V_a/V_b) \) obtained by the Eq. 4. The turnover rate \( (V_b) \), the formation rate \( (V_a) \) and the rate constant \( (K_s=V_a/A) \) of formation can be calculated from the turnover time, the pool size of lecithins \( (B) \), the ratio of \( V_a/V_b \) and the pool size of diacylglycerols \( (A; \mu\text{moles . g}^{-1} \text{ wet tissue}) \).

**RESULTS AND DISCUSSION**

The results are presented in Table 1. The ratio of \( V_a/V_b \) could be obtained with a good accuracy for saturated, dienoic and hexaenoic species. The data obtained for other species were found to be variable for unknown reasons. The results calculated for tetraenoic species could not be accepted as such, since radioactivity from precursors other than diacylglycerols could not be neglected as discussed previously (Moriya and Kanoh 1974).

Although heterogeneity of the lipid and cell components should be considered in the lung tissue, the results may indicate that dienoic species of lecithin can be formed mostly by de novo synthetic pathway (Kennedy 1961), but, in contrast, the formation of disaturated and hexaenoic species may be principally due to biosynthetic routes other than de novo pathway. The contribution of direct acylation pathway (Frosolono et al. 1971; Vereyken et al. 1972) and/or transacylation pathway (Akino et al. 1971) may explain the low \( V_a/V_b \) ratio observed in disaturated species. The low \( V_a/V_b \) ratios obtained for hexaenoic species may be explained by the participation of methylation pathway (Bremer and Greenberg 1961) as well as direct acylation pathway in the formation of this species.

The formation rate constant \( (K_s) \) suggests that saturated and oligoenic diacylglycerols may be utilized nearly equally by CDP-choline: diacylglycerol choline-phosphotransferase, that is, most of the lecithin species may be formed essentially according to the molecular composition of diacylglycerol pool. With rat liver microsomes, it has been indicated that choline phosphotransferase utilized
**TABLE 1. Calculation of metabolic turnover of lung lecithins**

<table>
<thead>
<tr>
<th>Lecithin species</th>
<th>$V_a/V_b^*$</th>
<th>Turnover time (min)</th>
<th>Rate constant of conversion of diacylglycerols (min$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total</td>
<td>0.76±9.50†</td>
<td>56±8.4</td>
<td>0.37</td>
</tr>
<tr>
<td>Saturated</td>
<td>0.18±0.03</td>
<td>39±7.9</td>
<td>0.40</td>
</tr>
<tr>
<td>Monoenoic</td>
<td>0.48±0.45</td>
<td>26±10.5</td>
<td>0.48</td>
</tr>
<tr>
<td>Dienoic</td>
<td>0.82±0.07</td>
<td>37±1.5</td>
<td>0.58</td>
</tr>
<tr>
<td>Tetraenoic</td>
<td>2.68±1.33</td>
<td>155±124</td>
<td>0.25</td>
</tr>
<tr>
<td>Hexaenoic</td>
<td>0.26±0.02</td>
<td>76±21</td>
<td>0.027</td>
</tr>
</tbody>
</table>

* $V_a$: The rate of formation of lecithins from diacylglycerols (µmoles·min$^{-1}$·g$^{-1}$).
* $V_b$: Turnover rate of lecithins (µmoles·min$^{-1}$·g$^{-1}$).
† Average values ±s.e. (three calculations).

without marked selectivity the diacylglycerol species differing in the degree of unsaturation, while CDP-ethanolamine: diacylglycerol ethanolamine-phosphotransferase utilized the hexaenoic species of diacylglycerol with a marked selectivity (Kanoh and Ohno 1975).

The low rate constant obtained for hexaenoic species of lecithin may be ascribed to the preferential utilization of hexaenoic diacylglycerol by ethanolamine-phosphotransferase in forming phosphatidylethanolamine in rat lung (Abe and Akino 1972).

The turnover time (less than one hr) of rat lung lecithins estimated in the early periods, as shown in Table 1, was markedly shorter than those determined in much later periods after administration of labeled precursors (Tierney et al. 1967; Spitzer and Norman 1971; Toshima et al. 1972; Abe et al. 1973). The reason for this discrepancy is difficult to explain, but the following points may be worthy of consideration:

1) The influx of radioactivity into lecithins cannot be neglected even in later periods after administration of labeled precursors as suggested by Young and Tierney (1972), and the half time or turnover time determined from the decrease of radioactivity in lecithins may become much longer than the real value.

2) The metabolic heterogeneity of the lung tissue may also be responsible for the discrepancy mentioned above. The very short turnover time shown in Table 1 may be a reflection of a rapid turnover of lecithins in the type II cells, as observed by Buckingham et al. (1966) while the longer half time determined from the decay of the phospholipids may be partly due to a slower turnover of the compounds in other types of cells in the lung tissue.

3) If the action of cholinephosphotransferase can be reversible in rat lung as well as in rat liver (Moriya and Kanoh 1972; Kanoh and Ohno 1973, 1975), this mechanism may also result in shorter turnover time of lecithins determined in the early periods. In this case the net formation of lecithins may be overestimated, since the disappearance of lecithin radioactivity by the conversion of lecithin to diacylglycerols would be negligible as compared with the influx of radioactivity into lecithin from diacylglycerols.
References


