The Mechanism of Acyl-Specific Phospholipid Remodeling by Tafazzin

Michael Schlame
Phospholipid

- Head group
- sn-1 Fatty acid
- sn-2 Fatty acid

Tafazzin

Chemical reactions and structures are shown involving phospholipids, fatty acids, and a protein called Tafazzin.
Tafazzin Reacts with < 1% of Mitochondrial Lipids

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<th>Mitochondria</th>
<th>dTAZ pmol/mg</th>
<th>PL nmol/mg</th>
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<tr>
<td>Fly WT</td>
<td>1.1±0.1</td>
<td>0.82±0.23</td>
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<td>Sf9 expressing dTAZ</td>
<td>20±3</td>
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<td>86±2</td>
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![Graph showing dTAZ (ng) vs. signal intensity](image1)

![Graph showing dTAZ (ng) vs. signal intensity](image2)
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![Graph showing changes in 14C-LPC and 14C-PC over time](image)

- Incubation time (min) vs. 14C-LPC and 14C-PC levels.
Tafazzin + PC/LPC Liposomes
Tafazzin + PC/LPC Liposomes

![Diagram](image)

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Tafazzin + PC/LPC Liposomes

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Supernatant: 82% [¹⁴C]PC
Liposomes: 100% PC
Tafazzin + PC/LPC Liposomes

![Graph showing the relationship between 14:0-LPC (nmol) and 18:2-18:2-PC (nmol).]
Tafazzin + PC/LPC Liposomes

31P NMR
Tafazzin Reacts only with Non-Bilayer Lipids

Phosphatidylethanolamine
Cardiolipin
Tafazzin Reacts only with Non-Bilayer Lipids

PC/LPC

PE/LPC

CL/LPC

Tafazzin activity
PC/LPC Micelles: Transacylation Activity Increases with the Number of Double Bonds and Decreases with the Chain Length
Tafazzin Transfers 14:0 Acyl Groups but does Not Achieve Complete 14:0 Exchange between PC & LPC

\[
14:0\text{-LPC} + 14:0\text{d-14:0d-PC} \rightarrow 14:0\text{-14:0d-PC} + 14:0\text{d-LPC}
\]
Conclusions I

- Tafazzin does not react with lipids in the bilayer state.

- Instead, it requires substrates to form assemblies with high positive or negative curvature, such as micelles or inverted hexagonal structures.

- Low lipid packing order promotes transacylation, presumably because it facilitates ideal mixing of PLs and LPLs.
Lysophospholipids

Phospholipids

Transacylations

$10^2$ Lysophospholipids

$10^3$ Phospholipids

$10^5$ Transacylations
Transition into H_{II} Phase Induces Acyl Specificity

18:1-CL

18:2-CL

18:1-CL+18:2-CL
Reconstitution of Mitochondrial Phospholipid Remodeling In Vitro

\[ \text{CL} : \text{MLCL} : \text{LPC} : \text{PC} \]
\[ 40 : 10 : 10 : 40 \]
Reconstitution of Mitochondrial Phospholipid Remodeling In Vitro

Diagram showing the reaction:

$\text{18:1-CL} \rightarrow \text{18:1} \rightarrow \text{18:1-LPC}$
$\downarrow \text{18:1} \downarrow \rightarrow \text{18:2-MLCL}$

Diagram showing the transacylation product (nmol):

- CL : MLCL : LPC : PC
- 40 : 10 : 10 : 40

Bar graph showing the transacylation product (nmol) with and without Ca$^{2+}$

- -Ca$^{2+}$
- +Ca$^{2+}$
Reconstitution of Mitochondrial Phospholipid Remodeling
In Vitro

$^{31}$P NMR

$^{31}$P Chemical shift (ppm)

+ Ca$^{2+}$

$L_{a}$

$H_{n}$
Reconstitution of Mitochondrial Phospholipid Remodeling In Vitro

\[
\begin{align*}
18:1-CL & \rightarrow 18:1-LPC \\
18:2-MLCL & \rightarrow 18:2-PC \\
\end{align*}
\]

\[
\begin{align*}
\text{Transacylation product (nmol):} & \\
\text{CL : MLCL : LPC : PC} & = 40 : 10 : 10 : 40 \\
\end{align*}
\]

\[
\begin{align*}
\text{CL : (18:2)_n-CL} & \\
\text{(18:1)_n-CL} & \\
\text{18:1-18:2-PC} & \\
\end{align*}
\]

- Ca\(^{2+}\)
+ Ca\(^{2+}\)
Conclusions II

• The acyl specificity of tafazzin depends on the lipid phase state.

• Native molecular species of cardiolipin can be created in vitro if tafazzin reacts with mitochondrial lipids in the hexagonal state.
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$^{31}$P NMR