The tripartite motif containing 24 acts as a novel coactivator of the constitutive active/androstane receptor

Yuichiro Kanno, Yuki Kure, Saori Kobayashi, Mariko Mizuno, Yumi Tsuchiya, Naoya Yamashita, Kiyomitsu Nemoto and Yoshio Inouye
Supplemental Material S1

<table>
<thead>
<tr>
<th>Time(min)</th>
<th>Buffer A</th>
<th>Buffer B</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.1</td>
<td>100.0</td>
<td>0.0</td>
</tr>
<tr>
<td>2.0</td>
<td>95.0</td>
<td>5.0</td>
</tr>
<tr>
<td>60.0</td>
<td>55.0</td>
<td>45.0</td>
</tr>
<tr>
<td>75.0</td>
<td>0.0</td>
<td>100.0</td>
</tr>
<tr>
<td>85.0</td>
<td>100.0</td>
<td>0.0</td>
</tr>
</tbody>
</table>

Buffer A; 2% Acetonitrile, 0.1%TFA
Buffer B; 80% Acetonitrile, 0.1%TFA

Figure legend of Supplemental material S1

LC-MS/MS systems at Oncomics Co., Lts as a custom service.

Ultrafiltration of co-immunoprecipitated proteins were used by ultrafiltration cartridge (AMICON UKTRA 0.5 3K, Millipore). Proteins were Cys. Alkylated and trypsin digested.

**Equipment and condition**

Nano-LC DiNa System (KYA TECH Corp.)

MS/MS TripleTOF® 5600(AB Sciex)

Time; 100min

**Data analysis**

ProteinPilotTM Software 4.5 (AB Sciex)

Sample type: Identification

Cys. Alkylation: Iodoacetamide

Digestion: Trypsin
HEK293 cells were transfected with expression plasmids for 3tag6 (empty) or 3tag6-TRIM24.

HEK293 cells were transfected with expression plasmids for shRNA expression plasmids for TRIM24-#1 (sh24-#1), TRIM24-#2 (sh24-#2) (0.1 µg), or GFP (shGFP).

After 48hr, whole cell lysates were resolved by SDS-PAGE, and proteins were detected by immunoblotting using antibodies against TRIM24 and Tubulin.