Retention time prediction and MRM validation reinforces the biomarker identification of LC-MS based phospholipidomics

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SUPPLEMENTARY FIGURES AND TABLES

Fig.S1 ECN-based RT predictive modeling of lysophospholipids. The predictive curve of LPC (A), LPE (B), LPG (C), LPI (D) and LPS (E) in negative ESI mode. X, represents carbon chain length, binomial curve fitting was used via y=a+bx+cx2, x represents relative carbon number (CN)), y represents relative RT.

Fig.S2 Predictive RT comparison between QSRR Automator and our established model. The actual and predicted RT of PC (A); The actual and predicted RT of PE (B); Actual RT refers to our established predictive RT. Predicted RT refers to QSRR predicted value.

Fig.S3 Identification characteristics for different subclass of phospholipids in the negative ESI mode.

Fig.S4 Lipids with same molecular formula have similar RT or separated RT. PC(18:0_20:5 and PC(18:2_20:3) of PC(38:5) can be separated at baseline(A); PC(18:1_18:3), PC(16:0_20:4) and PC(18:2_18:2) of PC(36:4) share the identical RTs(B).

Fig.S5 Lipids were robustly validated through two distinct MRM transitions. PC(16:0/18:1)(A), PE(16:0_18:1)(B), PG(16:0_18:1)(C), PI(18:0_18:1)(D), PS(18:0_18:1)(E) and PA(16:0_18:1)(F) were shown in both MRM ion pairs as above.

Table.S1 Retention time (RT) of deuterated internal standards.

Table.S2 Regressive curve of different subclass of phospholipids ($y=B0+B1x+B2x^2$).

Table.S3 Regressive curve of different subclass of lysophopholipids ($y=B0+B1x+B2x^2$).

Table.S4 MRM transition of lipids and deuterated internal standards.

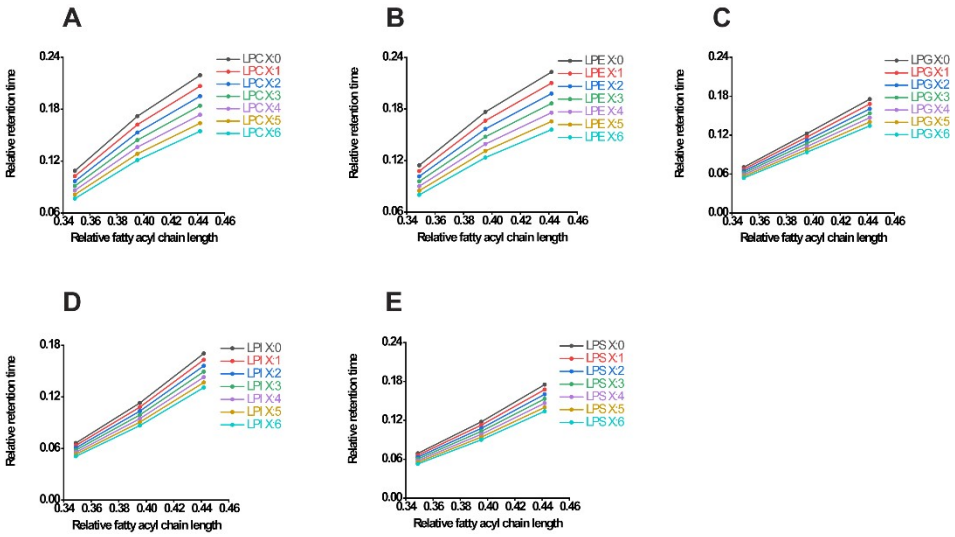
Table.S5 Expected and detected retention time of phospholipids.

Table.S6 Phospholipidomics of sorafenib resistant HCC cell lines.

Table.S7 Phospholipids with VIP>1 from OPLS-DA analysis of MHCC97H(1) and Hep3B(2).

Table.S8 Phospholipids with p (corr)>0.6 from OPLS-DA analysis of MHCC97H(1) and Hep3B(2).

Table.S9 Phospholipids with p (corr)<-0.5 from OPLS-DA analysis of MHCC97H(1) and Hep3B(2).



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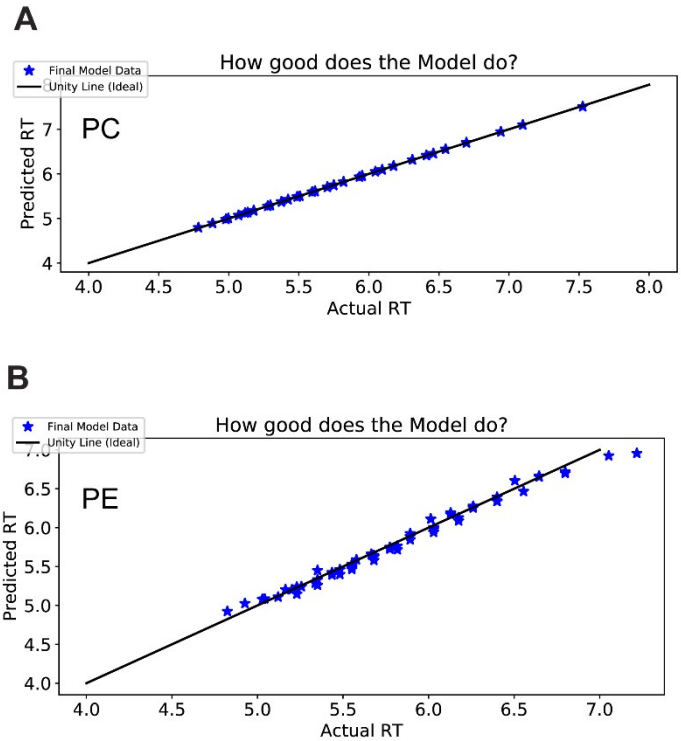

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Headgroup	Adduct	Characteristic fragment ions
PC	$[M+CH_3COO]^-$	$[FA1-H]^-$, $[FA2-H]^-$
PE	$[M-H]^-$	$[FA1-H]^-$, $[FA2-H]^-$
PG	$[M-H]^-$	$[FA1-H]^-$, $[FA2-H]^-$
PI	$[M-H]^-$	$[FA1-H]^-$, $[FA2-H]^-$
PS	$[M-H]^-$	$[FA1-H]^-$, $[FA2-H]^-$
FA1 and FA2 are fatty acids		

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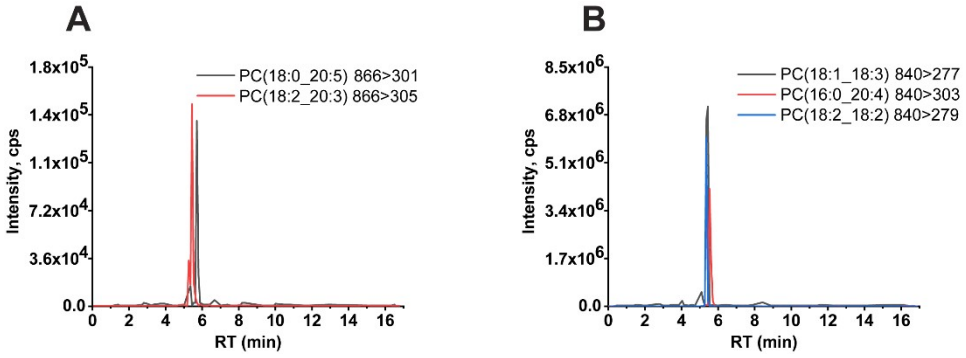

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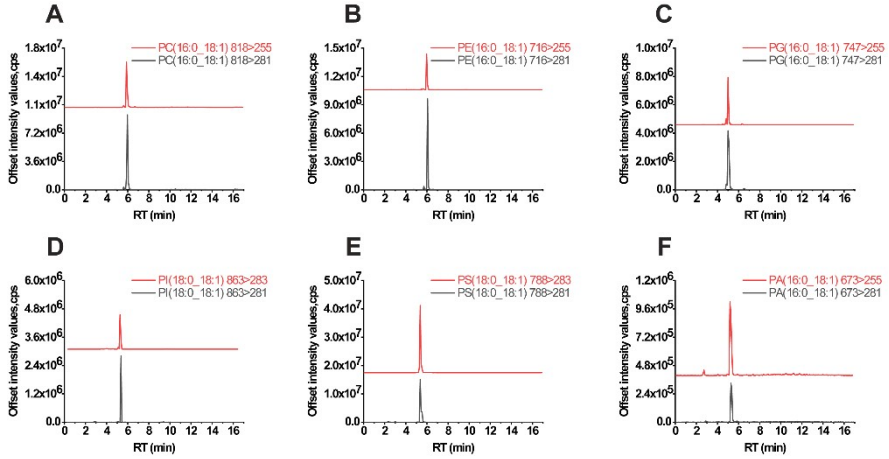

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