E07-09-0893 Arai

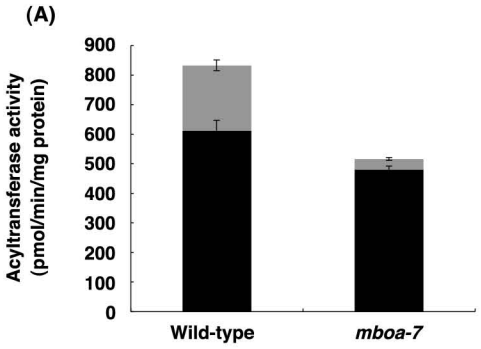
Supplementary Figure S1. Positional selectivity of MBOA-7. 40 μM of *sn*-1-lyso-2-acyl-lysoPI (see Materials & Methods) and 12.5 μM [^{14}C]arachidonoyl-CoA were used as an acyl acceptor and an acyl donor, respectively. After the *in vitro* acyltransferase assay, the lipids were extracted and separated by TLC as described in Materials & Methods. Significant radiolabeled PI was formed after the incubation with the microsomes of wild-type and *mboa-7* mutants (black plus gray bar). The resulting PI fractions were re-extracted from the TLC plates, treated with phospholipase A_2, and separated by TLC. Radiolabeled free fatty acid and lysoPI produced by the phospholipase A_2 treatment were indicated as gray bars and black bars, respectively.

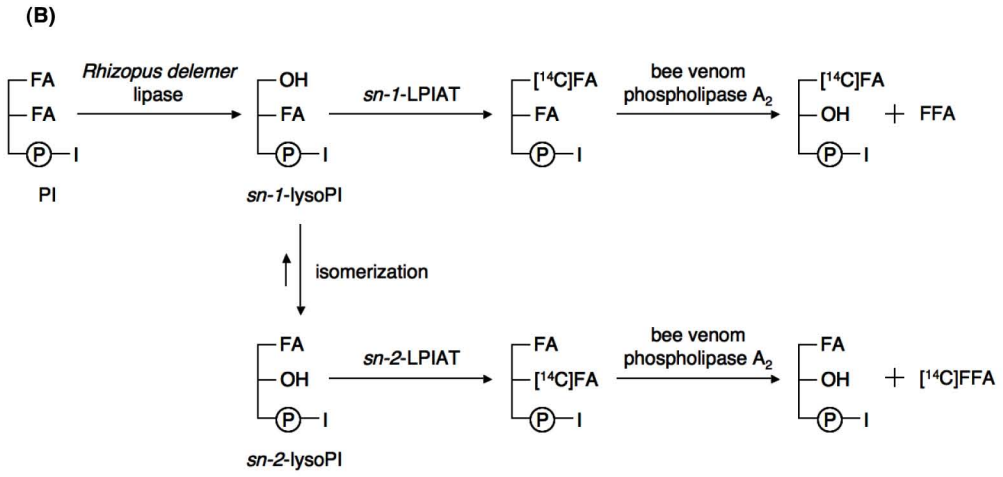
Supplementary Figure S2. ESI-MS/MS analysis of PI. (A) Negative ionization ESI-MS/MS spectra of PI molecular species of wild-type (upper) and *mboa-7* mutants (lower). PI molecular species were detected by diagnostic precursor scan of the dehydrated inositol phosphate fragment ion (m/z = -241). (B) Relative abundance of PI molecular species. Data are expressed as the percentage of intensity relative to an internal standard (16:0/16:0 PI). "a" refers to alkyl ether linkage.

Supplementary Figure S3. Knockdown of a human *mboa-7* homolog reduces incorporation of exogenous AA into PI. (A) Human *mboa-7* (*h-mboa-7*) mRNA was knocked down by siRNA oligonucleotides in HeLa cells as described under "Materials and Methods". Control siRNA having no significant homology to human gene

sequences (Ambion) was used as a negative control. The reduced expression of *h-mboa-7* was checked 72 h after siRNA transfection by real-time RT-PCR. Each bar represents the mean ± SEM of three independent experiments. *, $p < 0.05$. (B) Incorporation of [^{14}C]AA into phospholipids of HeLa cells. At 72 h after siRNA transfection, [^{14}C]AA was added to the medium, and the cells were incubated for 1 h. The cellular lipids were extracted to analyze the incorporation of radiolabeled fatty acids into phospholipids fractions. Each bar represents the mean ± SEM of three independent experiments. **, $p < 0.01$. (C) [^{14}C]Arachidonoyl-CoA:lysophospholipid acyltransferase activity of the membrane fractions of HeLa cells with different lysophospholipids as acyl acceptors. 12.5 μM [^{14}C]arachidonoyl-CoA and 40 μM each lysophospholipid were used as an acyl donor and acyl acceptor, respectively. Each bar represents the mean ± SEM of at least three independent experiments. **, $p < 0.01$.

Supplementary figure S1





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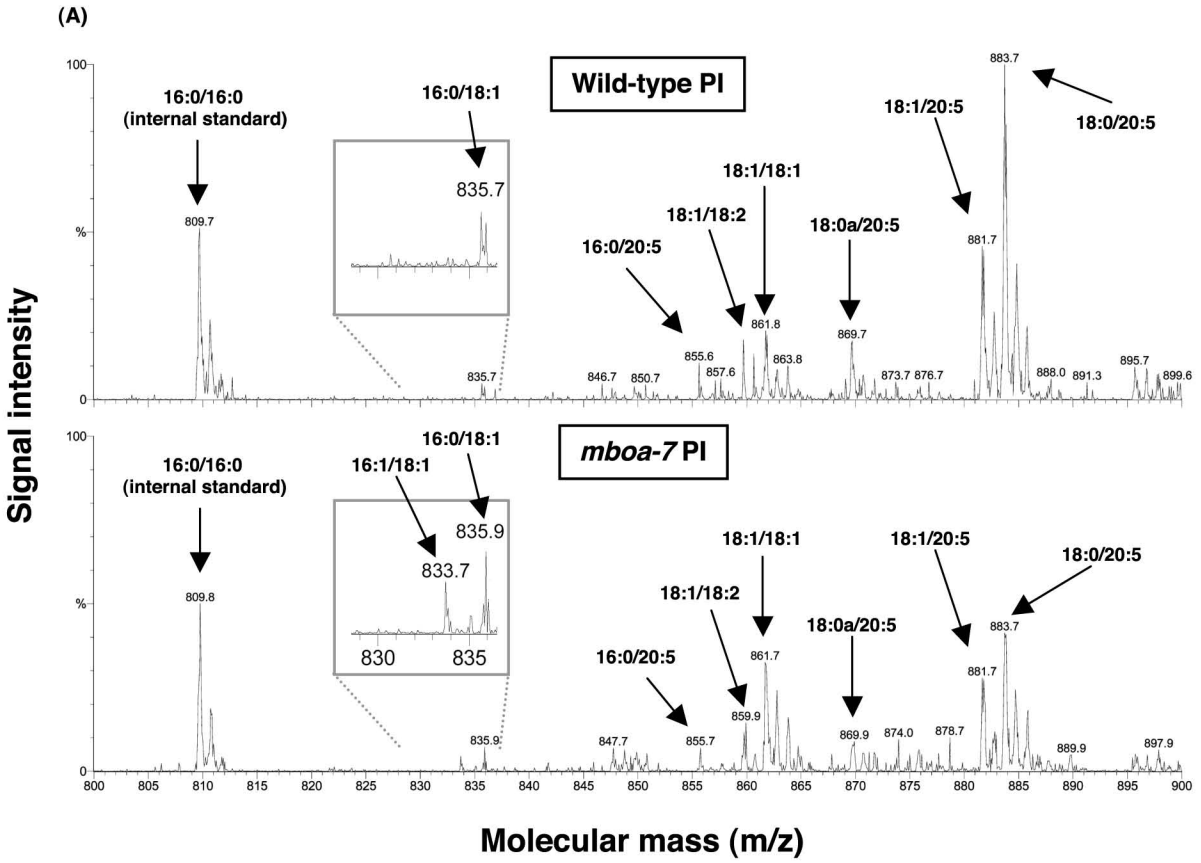
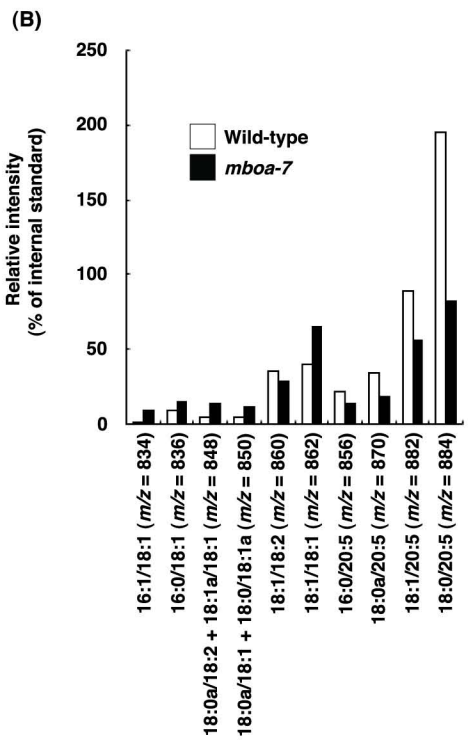
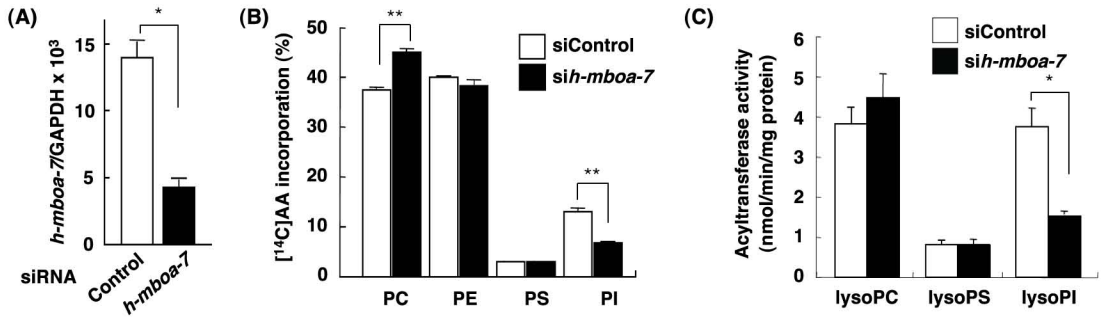
Supplementary figure S2





Supplementary figure S3



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