Supplemental Materials Molecular Biology of the Cell

Kong et al.

Supplemental Figure 1 A



В

Lag1 Lag2

500 bp -



Supplemental Figure 2

0



В

10 nM



С

10 nM

heated



Supplemental Figure 3



Supplemental Figure 1

(A) Chemical structure of pacFAceramide. N-(9-(3-pent-4-ynyl-3-H-diazirine-3-yl)-nonanoyl)-D-erythro-sphingosine is shown containing C18 sphingosine (top portion) and a C15 fatty acid modified with a diazirine ring at carbon 10 for UV cross-linking and a terminal alkyne group for Click chemistry modification. (B) RT-PCR using lag 1 and lag 2 primers based on predicted mRNA sequences in NCBI protein database.

Supplemental Figure 2

(A) Recovery of motility after incubation with myriocin. *Chlamydomonas* were treated with different concentrations of myriocin added to the medium as indicated. 15 h post-treatment, motility was tested (middle panel, left panel is control at time 0 h of incubation) and cell washed and resuspended in fresh medium without the addition of myriocin. Recovery of motility was assayed 48 h after incubation with fresh medium (right panel). Arrow indicates direction of light source. (B) Phase contrast image of *Chlamydomonas* after treatment with 10 nM myriocin for 15 h. (C) Trypan blue exclusion assay before and after treatment with 10 nM myriocin for 15 h. Right panel shows positive control obtained by heating *Chlamydomonas* at 60 °C for 15 min.

Supplemental Figure 3

Cilia on ependymal cells lacking neutral sphingomyelinase-2 are rescued by exogenous C24:1 ceramide. Z-scan orthogonal view of *in vitro* cultured wildtype and *fro/fro* mouse ependymal cells immunolabeled with anti-acetylated tubulin following incubation with or without C24:1 ceramide for 24 h. Scale bar, 5 µm.