Yamamoto et al. http://www.jcb.org/cgi/content/full/jcb.200510065/DC1

Supplemental materials and methods

LIVE/DEAD assay

Assay was performed following the manufacturer's (Invitrogen) instructions.

Western blots

 $20-40~\mu g$ of whole cell lysates were run on NuPAGE 10% Bis-Tris gels (Invitrogen) and transferred to polyvinylidene difluoride for 2 h. Blots were probed with anti-IRS-2 or anti-GFP overnight; they were then developed using an ECL detection kit (GE Healthcare).