**Table S1. Constructs used in this study**

|  |  |  |
| --- | --- | --- |
| REAGENT or RESOURCE | SOURCE | DESCRIPTIONS |
| Recombinant DNA | | |
| pAL-GST-LC3B(1-120aa) S3C | This study | Bacterial expression, 3C cleavage |
| pAL-GST-LC3B(1-120aa) S3C K42E/Q43E/L44E | This study |
| pAL-GST-LC3B(1-120aa) S3C R68E/R69E/R70E | This study |
| pAL-GST-GABARAP(1-116aa) K2C | This study |
| pAL-GST-GABARAP(1-116aa) K2C A39E/R40E/I41E | This study |
| pAL-GST-GABARAP(1-116aa) K2C R65E/K66E/R67E | This study |
| pGEX-6P1-GST-LC3B(1-120aa)-His6 S3C | This study |
| pGEX-6P1-GST-LC3B(1-120aa)-His6 K42C | This study |
| pGEX-6P1-GST-LC3B(1-120aa)-His6 L44C | This study |
| pGEX-6P1-GST-LC3B(1-120aa)-His6 R69C | This study |
| pGEX-6P1-GST-LC3B(1-120aa)-His6 R70C | This study |
| pAL-GST-GABARAP(1-116aa)-His6 K2C | This study |
| pAL-GST-GABARAP(1-116aa)-His6 V4C | This study |
| pAL-GST-GABARAP(1-116aa)-His6 V4C Nmut (M1E/K2E/E7A) | This study |
| pAL-GST-GABARAP(1-116aa)-His6 A39C | This study |
| pAL-GST-GABARAP(1-116aa)-His6 R40C | This study |
| pAL-GST-GABARAP(1-116aa)-His6 R65C | This study |
| pAL-GST-GABARAP(1-116aa)-His6 K66C | This study |
| pAL-GST-GABARAP(1-116aa)-His6 | This study |
| pAL-GST-GABARAP(10-116aa)-His6 ΔN9 | This study |
| pAL-GST-GABARAP(1-116aa)-His6 Nmut (M1E/K2E/E7A) | This study |
| pAL-GST-GABARAP(1-116aa) | This study |
| pAL-GST-GABARAP(10-116aa) ΔN9 | This study |
| pAL-GST-LC3B(1-120aa) | This study |
| pAL-GST-LC3B(12-120aa) ΔN11 | This study |
| pGEX-6P1-GST-ATG3 | Landajuela et al., 2016 |
| pBacPAK-his3-GST-ATG7 | This study | Sf9 insect cell expression, 3C cleavge |
| pFBDM- ATG7-ATG10-ATG12-StrepII2x-ATG5-ATG16L1 | This study | High Five insect cell expression, desthiobiotin elution |
| pMRX-ISU-hGABARAP | This study | Retrovirus infection |
| pMRX-ISU-hGABARAP D9 | This study |
| pMRX-ISU-hGABARAP ARI-EEE | This study |
| pMRX-ISU-hGABARAP Nmut(M1E/K2E/E7A) | This study |
| pMRX-ISU-hGABARAP Y49A L50A | This study |
| pMRX-IPU-EGFP-STX17TM | Tsuboyama et al., 2016 | Retrovirus infection |
| pQCXIH-ATG2A-FLAG | Tamura et al., 2017 | Expression in mammalian cells |

LANDAJUELA, A., HERVAS, J. H., ANTON, Z., MONTES, L. R., GIL, D., VALLE, M., RODRIGUEZ, J. F., GONI, F. M. & ALONSO, A. 2016. Lipid Geometry and Bilayer Curvature Modulate LC3/GABARAP-Mediated Model Autophagosomal Elongation. *Biophys J,* 110**,** 411-422.

TAMURA, N., NISHIMURA, T., SAKAMAKI, Y., KOYAMA-HONDA, I., YAMAMOTO, H. & MIZUSHIMA, N. 2017. Differential requirement for ATG2A domains for localization to autophagic membranes and lipid droplets. *FEBS Lett,* 591**,** 3819-3830.

TSUBOYAMA, K., KOYAMA-HONDA, I., SAKAMAKI, Y., KOIKE, M., MORISHITA, H. & MIZUSHIMA, N. 2016. The ATG conjugation systems are important for degradation of the inner autophagosomal membrane. *Science,* 354**,** 1036-1041.