## Effect of Serine/Threonine Kinases on Autophagy Mechanism

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Abstract : Autophagy is a degradation pathway, activating under stress conditions. It digests macromolecules, such as abnormal proteins and long-lived organelles by engulfing them and by subsequent delivery of the cargo to lysosomes. The members of the phospholipid-dependent serine/threonine kinases, involved in many signaling pathways, which are necessary for the regulation of cellular metabolic activation. Previous studies implicate that, serine/threonine kinases have crucial roles in the mechanism of many diseases depend on the activated and/or inactivated signaling pathway. Data indicates, the signaling pathways activated by serine/threonine kinases are also involved in activation of autophagy mechanism. However, the information about the effect of serine/threonine kinases on autophagy mechanism and the roles of these effects in disease formation is limited. In this study, we investigated the effect of activated serine/threonine kinases on autophagic pathway. We performed a commonly used autophagy technique, GFP-LC3 dot formation and by using microscopy analyses, we evaluated promotion and/or inhibition of autophagy in serine/threonine kinase-overexpressed fibroblasts as well as cancer cells. In addition, we carried out confocal microscopy analyses and examined autophagic flux by utilizing the differential pH sensitivities of RFP and GFP in mRFP-GFP-LC3 probe. Based on the shRNA-library based screening, we identified autophagyrelated proteins affected by serine/threonine kinases. We further studied the involvement of serine/threonine kinases on the molecular mechanism of newly identified autophagy proteins and found that, autophagic pathway is indirectly controlled by serine/threonine kinases via specific autophagic proteins. Our data indicate the molecular connection between two critical cellular mechanisms, which have important roles in the formation of many disease pathologies, particularly cancer. This project is supported by TUBITAK-1001-Scientific and Technological Research Projects Funding Program, Project No: 114Z836. Keywords : autophagy, GFP-LC3 dot formation assay, serine/threonine kinases, shRNA-library screening

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