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Decreased Autophagy Contributes to Senescence Induction in HS68 Cells

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Abstract : Ageing is associated with an increased risk of diseases such as cancer, and neurodegenerative disorders. Increased autophagy delays ageing and extends longevity. In this study, we investigated the role of autophagy in longevity using human foreskin fibroblast HS68 cells, in which a senescence-like growth arrest can be induced. In particular, cellular senescence is manifested by the irreversible cell cycle arrest, and may contribute to the ageing of organisms. The senescence state was measured with staining for senescence-associated β-galactosidase (SA-β-gal) activity that represents a sensitive and reliable marker to quantify senescent cells. We detected a significantly increased percentage (%) of SA-β-gal positive cells in HS68 cultures at passage 40 (63%) when compared with younger ones at passage 15 (0.5%). As expected, HS68 cells at passage 40 exhibited much lower proliferation rate than cells at passage 15. The basal levels of LC3 were measured by immunoblotting showing a comparison of LC3-I and LC3-II levels at 3 age-points in serially passaged HS68 cells. LC3-II/LC3-I ratio at different passage levels relative to β-actin levels of each band confirmed that cells at passage 34 showed lower conversion of non-autophagic LC3-I to autophagic LC3-II than the cells at passage 16. Furthermore, Cyto-ID autophagy assay also revealed that late passage cells showed lower autophagy than the early passage cells. Together, our findings suggest that senescence induction might be associated with decreased autophagy.

Keywords : ageing, autophagy, senescence, HS68
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