Development of a screening system for the compounds that induce the cell cycle activity in mammalian cardiomyocytes

Risa Matsui¹, Shoki Fukuyama¹, Yusuke Kametani¹, Yuriko Wada¹, Kotaro Matsumoto¹, Makiko Maeda², Masanori Obana¹, Yasushi Fujio¹

¹Laboratory of Clinical Science and Biomedicine., Grad. Sch. Pharm., Osaka Univ., ²Education and Research Project for Pharm.D., Grad. Sch. Pharm., Osaka Univ.

【Background and Objective】

Since mammalian cardiomyocytes exit from cell cycle immediately after birth, the regenerative activity is limited in adult mammalian hearts. The aim of this study is to construct a screening system of the compounds that induce the cell cycle activity in mammalian cardiomyocytes, using Geminin-mAG1, a Fluorescent Ubiquitination-based Cell Cycle Indicator (FUCCI) that is stabilized in S/G2/M phases.

【Methods and Results】

We generated adenovirus vector expressing the FUCCI and infected the vector in neonatal rat cardiomyocytes. The frequency of FUCCI-positive cardiomyocytes increased in a FBS concentration dependent manner, concomitant with Ki-67-positive cells. Importantly, the cells transfected with miR-294, which was reported to be involved in cardiomyocyte proliferation, exhibited increased frequency of FUCCI-positive cells. In the presence of the proteasome inhibitor MG-132 that stabilizes FUCCI, more than 90% of the cells were positively identified, which is the limitation of this screening system.

【Conclusion】

We constructed a novel screening system for the compounds that induce the cell cycle activity in cardiomyocytes, though false positive samples cannot be completely excluded.