While rapamycin was discovered as a fungicide in 1975, it is now famous for its ability to suppress mammalian cell aging and to extend lifespan in mice. Rapamycin is an oral immunomodulatory agent approved to prevent rejection in human organ transplantation. It also exerts an antitumor activity as well as an anti-restenosis activity, when used in drug-eluting stents. In addition, rapamycin has neuroprotective effects in traumatic brain injury models, with increased neuron survival and recovery of brain functions. Nevertheless, there has been a practical limitation in performing physiologic studies with rapamycin, because rapamycin has very poor water solubility and bioavailability. To maximize the research and clinical application of rapamycin, it is advantageous to dissolve rapamycin in a physiologic and isotonic solution.

To overcome this limitation (poor water solubility), we solubilized rapamycin in isotonic buffer using reconstituted high-density lipoprotein containing V156K-apoA-I (V156K-rHDL). Apolipoprotein (apo) A-I is a principal protein component of HDL and is known to perform a beneficial role in the reverse cholesterol transport pathway through its antioxidant and antiinflammatory activities. HDL have atheroprotective effects with potent antioxidant, antiinflammatory, anti-diabetic activities. Recently, we showed that rHDL containing V156K-apoA-I, had a potent antioxidant activity and was more resistant to glycation, with more enhanced tissue regeneration activity as a HDL-therapy. More resistance to glycation is advantageous to develop a protein-based drug for both anti-atherosclerotic and anti-diabetic agents. These results suggest that V156K-rHDL could be applied to facilitate facultative regeneration in aging-related complications.

There has been several nanobiotechnology applications of rHDL to encapsulate doxorubicin, amphotericin and adenovirus. Our research group recently reported that the efficiency of viral gene delivery and the stability of the adenovirus (Ad) were significantly enhanced when Ad is incorporated with rHDL containing WT- or V156K-apoA-I in cellular and zebrafish models. The report raised the feasibility that rHDL can be used as a formulated vehicle for viral and gene/drug delivery.

We showed a new method to solubilize rapamycin for better delivery across several barriers, such as blood-brain barrier and skin barrier. Rapamycin (final 0.1 mg/mL) was solubilized in rHDL containing either wild-type (WT) or V156K (1 mg/mL of protein). V156K-rHDL containing rapamycin (V156K-rapa-rHDL) had a slightly larger particle size than WT-rHDL containing rapamycin (WT-rapa-rHDL). V156K-rapa-rHDL had enhanced antioxidant ability and anti-atherosclerotic activity. Treatment of V156K-rapa-rHDL resulted in attenuated senescence in human cells, with increased cell survival compared with WT-rapa-rHDL or rHDL alone, indicating that V156K-rapa-rHDL has various applications for suppression of cellular senescence and tissue regeneration activities in the zebrafish model. Incorporation of rapamycin into rHDL containing apoA-I, especially its V156K variant, can promote cell survival and suppress senescence through enhanced antioxidant ability. V156K-apoA-I has been developed by site-directed mutagenesis by our research group and has been shown to have more potent antioxidant and anti-inflammatory activities in vivo and in vitro, as well as in vivo anti-atherosclerotic effect.

In conclusion, the enhanced solubility of rapamycin in rHDL particles may be applicable to the development of numerous types of therapeutics. Because cardiovascular disease and type 2 diabetes are age-related diseases, chronic metabolic disease progression with senescence may be linked to target of rapamycin (TOR) signaling. Rapamycin and rHDL possess independent therapeutic activities, and the beneficial activities of rHDL were not impaired by incorporation of rapamycin. New agents can likely be developed from rapa-rHDL to treat aging, cardiovascular and brain disease, as HDL can penetrate the blood-brain barrier. These same protocols can be applied to solubilize other hydrophobic drugs for a wide range of therapeutic applications.

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