Synopsis of Original Research Paper

Control of secretion polarity of bioactive protein after gene delivery to the skin

Yoshinobu Takakura

Graduate School of Pharmaceutical Sciences, Kyoto University

Epithelial cells, generally displaying cell polarity, are considered to be good targets for gene therapy because of their distribution of enormous surface area of various tissues and organs in animal bodies. Polarized epithelial cells possess two functionally and compositionally distinct cell surface domains that are separated from each other by tight junctions. They play fundamental roles in the specialized vectorial transport and secretion of proteins in tissue and organs.

Newly synthesized domain-specific membrane proteins are conveyed through the endoplasmic reticulum (ER)-Golgi apparatus to the trans-Golgi network (TGN), and packaged into transport carriers for membrane integration. The destination of the membrane proteins seems to be determined by interrelation between some signal sequences on the protein and various cellular components playing roles in sorting and trafficking. Secretory proteins are also considered to be transported in the same manner as the membrane proteins. In spite of many investigations of the issue, the mechanisms regulating apical or basolateral sorting of secretory proteins remain elusive.

The green fluorescent protein (GFP) of the jellyfish Aequorea victoria and its variants are widely used in cell imaging applications to reveal the location of proteins. Results from those applications are providing new insights into protein function and cellular processes in the complex environment of the cell. The GFP-tagging technology is useful for directly investigating the intracellular localization, movement and fate of secretory proteins of interest in living cells.

We have recently examined the mode of secretion polarity of interferon (IFN)-β expressed exogenously in several epithelial cell lines. The secretion of constitutive IFN-β from the stable transformants was apparently unpolarized. Meanwhile, intriguingly, IFN-β transiently expressed by gene transfection was predominantly secreted from the cell membrane side to which the transfection or the induction was carried out. In this study, we have investigated the subcellular localization of the cytokine using GFP-tagged IFN-β under confocal laser scanning microscopy (CLSM). Our results suggested that IFNs expressed stably and transiently are transported via different post-TGN vesicles. The same secretory protein, at least as far as IFN is concerned, can be sorted to the apical or basolateral membrane side depending on the gene expression strategy, which deems to be regulated at the post-TGN stages.