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申請者（ふりがな）	久原 麻那 （ くはら まな ）
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発表者（※学会発表の場合のみ記載、共同発表者の氏名も記載すること）	久原 麻那, 丸亀 裕貴, Wang Jinyun, 板倉 英祐, Xie Kun, 矢野 敏史, 原 太一
発表題目（※学会発表の場合のみ記載）	6-MSITC Enhances Autophagy and Lysosomal Function via Activation of TFEB and Cross-talk with Narf2

発表の概要と成果（抄録を公開している URL がある場合、「概要・成果」を記載した上で、URL を末尾に記してください。また、抄録 PDF は別途ご提出ください。なお、抄録 PDF は Web 上には公開されません。）

Objective: Wasabi contains a unique isothiocyanate, 6-(methylsulfinyl)hexylisothiocyanate (6-MSITC), which is expected to have health benefits such as anticancer activity and prevention of lifestyle-related diseases. It has been shown that autophagy, a recycling system of intracellular components, functions as an important defense system against such diseases and aging. We have shown that 6-MSITC activates autophagy. In this study, we aimed to elucidate the mechanism of autophagy regulation by 6-MSITC from horseradish and the role of autophagy in the maintenance of health at the cellular level.

Materials & Methods: Autophagy flux assay was performed using cells stably expressing a fluorescent probe (GFP-LC3-RFP-LC3ΔG, tandem fluorescent-tagged LC3) that can measure autophagy flux, and fluorescence intensity quantification using FACS and fluorescence microscopy and Autophagic flux (degradation activity by autophagy) activity was quantitatively evaluated by microscopic observation. cells stably expressing TFEB-GFP were used to observe the subcellular localization of TFEB, which regulates lysosome biogenesis,

using fluorescence microscopy. Effects on lysosome number and activity (acidification) were evaluated by cell staining using LysoTracker and observation and quantitative analysis using FACS and fluorescence microscopy. The effects of TFEB and Nrf2 on downstream gene and related protein expression were analyzed by qPCR and Western blotting.

Results & Discussion: In this study, we focused on the role of lysosomes in the autophagy process and analyzed how 6-(methylsulfinyl)hexylisothiocyanate (6-MSITC), a functional component derived from horseradish, acts in cells. In particular, we predicted that the lysosomal damage response mechanism is involved in the intracellular sensing of polyphenols and focused on the transcription factor TFEB (master regulator of lysosomal biosynthesis), a key factor in this process. As a result, we found that 6-MSITC promotes nuclear translocation of TFEB, increases expression of TFEB target genes, and enhances lysosomal function. In TFEB knockout cells, 6-MSITC-induced activation of autophagy and enhancement of lysosomal function were attenuated, confirming the involvement of TFEB. Notably, 6-MSITC requires not only TFEB but also Nrf2 to induce autophagy. These results suggest that the lysosomal biosynthesis pathway and the antioxidant response system may regulate autophagy through crosstalk, suggesting that lysosomes may play an important role in the regulation of cellular stress response mechanisms by 6-MSITC. This crosstalk may allow 6-MSITC to provide multifaceted cellular stress protection and contribute to the extension of healthy life span and improvement of quality of life.