培養肝細胞におけるオートファゴゾーム形成膜の免疫電顕による研究

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Immunocytochemical Study of the Surrounding Envelope of Autophagic Vacuoles in Cultured Rat Hepatocytes

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ABSTRACT By the use of erectron immunoperoxidase cytochemistry at the ultrastructural level, the relationship of the surrounding sac of the autophagic vacuoles to the different cytomembranes was studied. When the endoplasmic reticulum was completely stained for microsomal carboxyesterase E1, the enzyme was not found to be labeled in the developed envelopes forming autophagic vacuoles. The autophagic envelope at the formative stages was also devoid of albumin which intensely stained Golgi cisternae. However, although it was rare, the endoplasmic reticulum showed an electron－lucent region like an early autophagic envelope in its cisternae which was lacking in carboxyesterase E1. In addition, deeply curving swelled cisternae where carboxyesterase E1 was found at the edges were occasionally encountered. These observations suggest that the segregating membranes arise from an endoplasmic reticulum and the structural characteristics of the endoplasmc membranes change at very early stages of formation of autophagic vacuoles. Acid phosphatase, a lysosomal marker enzyme, began to be localized on sections of the double membranes of newly created autophagic vacuoles. The enzyme spread all along the limiting membranes of the autophagic vacuoles, while, at the same time, the double membranes were converted into a single membrane. A lysosomal membrane glycoprotein (LGP 107) was also localized on the surrounding envelope of autophagic vacuoles in a fashion similar to that of acid phosphatase. Lysosomal hydrolases seem to play some role in the conversion of double limiting membranes

into a single limiting membrane.

抄録　免疫電顕法を用いて，オートファゴゾーム膜の細胞内膜からの派生について調べた。ゴルヂ膜をアルブミンで染色した時，オートファゴゾーム膜形成過程に本蛋白が染色されることはなかった。オートファゴゾーム形成膜は，小胞体構成成分であるエステラーゼによっても染色されなかった。しかし，本蛋白がオートファゴゾーム形成初期段階の小胞体膜には欠損している像が見い出され，小胞体膜がオートファゴゾーム膜へ変化する初期段階で膜構造が変化することが示唆された。次に，ライソゾーム膜構成成分であるアシドホスファターゼやLGP 107でファゴゾーム膜を染色すると，両蛋白は二重膜が一重膜に変化する前からファゴゾーム膜に検出された。この事からオートファゴゾーム膜の成熟にライソゾームが関与していることが示唆された。

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