

## ラット肝リソゾーム膜に存在する主要な 糖蛋白質の精製と性質

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### Purification and Characterization of a Major Glycoprotein in Rat Liver Lysosomal Membrane

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**ABSTRACT** A major lysosomal membrane glycoprotein (LGP 170) which has an apparent molecular weight ( $M_r$ ) of 107 kilodaltons (kDa) was purified from rat liver by a simple method with a yield of 1 mg / 87 g wet weight of liver. The purification procedures include ; preparation of tritosomal membranes of triton-filled lysosomes (tritosomes), extraction of tritosomal membranes by Lubrol PX, wheat germ agglutinin (WGA) - Sepharose affinity chromatography, and monoclonal antibody - Sepharose affinity chromatography. The quantitative immunoblot analysis indicated that LGP 107 represents 6.2% of the total protein of tritosomal membranes. The isoelectric point of the purified glycoprotein was 2.7, and it moved toward neutral pH after sialidase treatment, with its molecular weight decreased by about 10 kDa. LGP 107 contained 52% carbohydrates, and the carbohydrate moiety was composed of Fuc, Man, Gal, GlcNAc and sialic acid in a molar ratio of 7.2 : 68.2 : 40.6 : 63.0 : 32.3, respectively, indicating that LGP 107 was highly glycosylated with *N*-linked complex-type oligosaccharide chains. Out of the *N*-linked glycans released from the glycoprotein by hydrazinolysis / *N*-reacetylation, about 70% was sialylated. Anion exchange and reverse-phase high performance liquid chromatography analysis on the structure of *N*-glycans revealed that a disialyl biantennary form is a major component in the oligosaccharide chains of LGP 107.

抄録 ラット肝ライソゾーム膜に存在する主要な糖蛋白質 (LGP 107) を精製し, その性質を調べた。LGP 107 はトライソーム膜蛋白質の 6.2% を占める主要な構成成分であった。LGP 107 はその分子内にある多量のシアル酸により, 等電点 2.7 を示した。LGP 107

は全重量の52%の糖質を含有していた。糖組成を分析すると LGP 107 の糖鎖は Asn に結合した複合型糖鎖であり、その70%はシアロ化されていた。さらに HPLC を用いて糖鎖構造を調べると、disialyl biantennary 構造が、主要な糖鎖であった。

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