Identification and Characterization of LAMP-1 as an Activation-

dependent Platelet Surface Glycoprotein*

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Platelets normally circulate in a quiescent state. When activated, they undergo biochemical and morphological changes which greatly alter their function and contribute to their role in thrombosis and hemostasis. We have identified, cloned, and sequenced a cDNA from a human umbilical vein endothelial cell library that encodes a 110-kDa integral membrane protein. This protein is present on the surface of activated but not resting platelets and has previously been identified as lysosomal-associated membrane protein 1 (LAMP-1). Half-maximal surface expression of platelet LAMP-1 was induced by concentrations of thrombin that resulted in lysosome enzyme release, not α-, or dense granule release. Also consistent with lysosome enzyme studies, there was little surface expression of LAMP-1 in response to the weak agonists ADP and epinephrine.

In addition, sucrose density gradient fractionation of platelet granules showed colocalization of LAMP-1 with the lysosomal enzyme, β-galactosidase, and not with markers of α- or dense granules. While we found virtually no LAMP-1 on the resting platelet surface (0–90 molecules/cell), we estimated a mean of 1175 LAMP-1 molecules on the thrombin-activated platelet surface. The translocation of this heavily glycosylated protein to the platelet surface upon stimulation may play a role in the adhesive, prothrombic nature of these cells.

Upon contact with any of a variety of stimuli, platelets change shape, release their granular components, and aggregate with one another to form the primary thrombus. As part of this change from a resting to a prothrombotic, adhesive phenotype, the protein profile of the cell surface undergoes a transformation. This is accomplished by several incompletely understood mechanisms. For example, a calcium-dependent conformational change occurs in the glycoprotein IIb/IIIa integrin complex, resulting in the acquisition of functional capacity as a fibrinogen receptor, which then mediates platelet aggregation (Bennet and Vilaire, 1979). PADGEM (O-90 molecules/cell), we estimated a mean of 1175 LAMP-1 molecules on the thrombin-activated platelet surface. The translocation of this heavily glycosylated protein to the platelet surface upon stimulation may play a role in the adhesive, prothrombic nature of these cells.

EXPERIMENTAL PROCEDURES

Two human umbilical vein endothelial cell cDNA libraries, one in LMT11, the other in LMT10, were kindly provided by Dr. M. Chao (Cornell University Medical College). The libraries contained about 10^6 independent recombinants and were amplified once before screening. The inserts were cloned into the EcoRI site of the vector. The LMT10 library was size selected (>1 kb cDNAs).

A rabbit antiserum with broad anti-platelet reactivity was a gift of Dr. J. Barnwell (New York University). Murine monoclonal anti-LAMP-1 antibody (H5G11) and rabbit polyclonal anti-LAMP-1 serum were gifts of Dr. J. T. August (Johns Hopkins University), and monoclonal anti-PADGEM (AC1.2) of Dr. B. Furie (Tufts University). Rabbit polyclonal anti-β-galactosidase antibody was purchased from Cappel. FITC-conjugated goat anti-rabbit and anti-mouse IgG were purchased from TACO ( Burlingame, CA). Alkaline phosphatase-conjugated goat anti-rabbit IgG was purchased from Bio-Rad, as were the colorimetric substrates, 5-bromo-4-chloro-3-indolyl phosphate p-toluidine salt and p-nitro blue tetrazolium chloride.

Radioactive nucleotides, [α-32P]dATP, 3000 Ci/mM, and [35S] dCTP, >1000 Ci/mM, 125I-NaI (100 Ci/mM), and 5-hydroxy(aside

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1 The abbreviations used are: LAMP-1, lysosomal-associated membrane protein 1; FITC, fluorescein isothiocyanate; SDS, sodium dodecyl sulfate; Heps, 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid; ELISA, enzyme-linked immunoabsorbent assay; TSP, thrombospondin; bp, base pairs; HBSS, Hanks' balanced salt solution.
were purchased from United States Biochemical Company (Cleveland, OH). Isopropyl-β-D-thiogalactopyranoside, ADP, and indomethacin were purchased from Sigma. D-Phenylalanyl-L-propyl-L-arginine chloromethyl ketone was from Calbiochem.

Library Screening

Two million AGT11 recombinants were plated in the primary library screen. After 5–7 d of infection, the bacteria were overlaid with nitrocellulose previously soaked in 10 mM isopropyl-β-D-thiogalactopyranoside (Young and Davis, 1983). The filters were screened immunologically with the anti-platelet IgG, and positive clones were identified using an alkaline phosphatase colorimetric system (Blake et al., 1984). The screening antisera and second antibody were pre-cleared with a freeze-thaw lysate of Y1090 bacteria.

Screening of the AGT10 library was by hybridization (Benton and Davis, 1977), using plaque-purified, immunologically identified, cloned cDNA inserts as probes. Two million plaques were plated for the primary screen, and transferred to nitrocellulose. After washing (final wash in 0.2 × SSC (0.15 M NaCl, 0.015 M sodium citrate, pH 7.0), 0.1% SDS, 0.1% sodium pyrophosphate at 50 °C for 15 min) the filters were overlaid on Kodak X-omat film and autoradiographs were obtained.

Affinity Purification of Anti-fusion Protein Antibody

Escherichia coli strain Y1090 were infected with plaque purified phage at a high density. Fusion protein production was induced and the bacterial lawn overlaid with nitrocellulose as above. The nitrocellulose was then incubated in a solution of 20 mM Tris, 0.15 M NaCl, 0.05% Tween 20, 5% nonfat dry milk (Carnation), pH 7.4, overnight at 22 °C. Small pieces of the filter were incubated with the screening IgG for 2 h at 22 °C, rotating. The filters were washed and overnight at 22 °C. Small pieces of the filter were incubated with the screening IgG for 2 h at 22 °C, rotating. The filters were washed and overnight at 22 °C. Small pieces of the filter were incubated with the screening IgG for 2 h at 22 °C, rotating. The filters were washed and overnight at 22 °C.

DNA Sequencing
cDNA inserts were subcloned into M13mp18 or M13mp19 and both strands sequenced as far as possible using the dideoxy-chain termination method (generally ~350–500 bp) (Sanger et al., 1977). T4 DNA polymerase (Sequenase) and universal sequencing primers were purchased from United States Biochemical Company (Cleveland, OH). Oligonucleotides of 21 bp were then synthesized based on a previous sequence and used to prime subsequent reactions. These custom oligonucleotides were purchased from Genosys (Houston, TX) or were obtained from the Cornell Medical College Core DNA synthesis facility. In this way, >90% of the sequence was determined in both orientations.

Northern Blot

Total cellular RNA was isolated from human umbilical vein endothelial cells by the acid/guanidinium/phenol/chloroform method (Chomczynski and Sacchi, 1987). Twenty µg were electrophoresed in 1% agarose gels containing 2% formaldehyde. After transfer to nitrocellulose, blots were probed with [32P]labeled cloned insert DNA, labeled to high specific activities. Blots were hybridized overnight at 50 °C and then washed stringently, ending at 0.2 × SSC, 0.1% SDS, 0.1% sodium pyrophosphate, at 42 °C.

Plaque Isolation

Platelets were prepared from 20 ml of fresh blood in 2.7 ml of acid citrate dextrose (38 mM citric acid, 75 mM sodium citrate, 136 mM dextrose). After a low speed spin (200 × g, 15 min), the top half of platelet-rich plasma was applied to a 25-cm column of Sepharose 2B pre-equilibrated with buffer solution. The column was washed with 2.7 ml of 0.9% NaCl, 0.075 M Na2HP04, 5.5 mM glucose, pH 7.4 containing 0.2% bovine serum albumin. The platelet peak was pooled and, in some cases, recollected to 1 ml.
sample buffer was added to the supernatants. The samples were boiled, and 1 μl of the lysate was electrophoresed on a gradient (10-15%) polyacrylamide gel using the Phastgel system (Pharmacia). Three ng of purified human LAMP-1 were run as a control. Proteins were transferred to nitrocellulose by capillary action (Smith, 1989), and the filters were incubated for 18 h in a solution of TBS containing rabbit anti-LAMP-1 IgG and the filters were incubated for 18 h in a solution of TBS containing 0.05% Tween, and 5% Carnation instant nonfat dry milk. Blots were then incubated in TBS/Tween containing rabbit anti-LAMP-1 IgG or non-immune IgG for 1 h at 22 °C. After washing in TBS/Tween, they were incubated with alkaline phosphatase-conjugated goat anti-rabbit IgG for 20 min. The blots were washed and developed using the 5-bromo-4-chloro-3-indolyl phosphate p-toluidine salt/p-nitroblue tetrazolium chloride color development reagent system (Blake et al., 1984).

RESULTS

Identification and Analysis of the Activation-dependent Clone—Approximately 2 × 10⁶ XGT11 recombinants from a human umbilical vein endothelial cell cDNA library were screened with a broadly reactive polyclonal anti-platelet antibody. This library was chosen for screening because platelets have little mRNA and because endothelial cells have many proteins in common with platelets. The initial screen yielded 40 positives. At the end of the tertiary screen, there were five highly positive clones: three cross-hybridized and were pursued further. The other two have not been completely characterized.

Flow cytometric analysis of platelets using IgGs affinity purified on immobilized recombinant fusion proteins identified a cDNA encoding an antigen expressed preferentially on thrombin-activated platelets. As shown in the flow cytographs in Fig. 1, while the screening IgG reacted with both resting and activated platelets (panel A), the affinity purified IgG bound only to activated platelets (panel B). Control (non-immune) rabbit IgG did not react with fusion protein or with platelet surfaces. Thus, this cDNA encodes a peptide epitope unique to the surface of activated platelets.

The cDNA was estimated to be approximately 400 bp based on agarose gel analysis and by the size of the β-galactosidase fusion protein, determined by Western blot (data not shown). Nucleotide sequence analysis showed it to be different from PADGEM (GMP140), GPIIIb/IIIa, fibrinogen, TSP, fibronectin, and von Willebrand factor. To identify full-length cDNAs, a size-selected human umbilical vein endothelial cell cDNA library in λgt11 was screened with this insert DNA. Several overlapping clones were identified, mapped, and sequenced. The largest was 2.2 kilobases and included the 3' end, identified by a consensus polyadenylation signal (AAUAAA) 19 bp 5' to a poly(A) tail (A0). An analysis of the nucleotide and deduced protein sequence revealed identity (98% homology) to another recently cloned protein, human lysosomal associated membrane protein 1 (Chen et al., 1988; Fukuda et al., 1988). A comparison of the nucleotide sequence we obtained with that of LAMP-1 revealed a single difference in the coding region (nucleotide 367), but the amino acid is conserved. In the untranslated 5' region, there were a number of differences, including a 25-base pair insertion at nucleotide 1850, and the presence of a poly(A) tail. LAMP-1 was reported to be an integral membrane protein of the lysosome (Chen et al., 1988). Nucleotide sequence predicted that LAMP-1 is transcribed as a 416-amino acid protein. There is a 27-amino acid sequence consistent with a classic signal peptide at the amino-terminus. LAMP-1 consists of two homologous domains, each containing 4 evenly spaced (36-38 residues) cysteines, separated by a Pro-Ser-rich IgA-like hinge region. There is a 24-amino acid 5' hydrophobic region, consistent with a transmembrane domain and a short 11-amino acid cytoplasmic tail. The predicted extracytoplasmic (intralysosomal) domain contains 18 consensus sites for N-linked glycosylation. The 3'-untranslated region is remarkable for a series of repeats that are extremely guanosine-rich.

Northern blot analysis of HUVEC total RNA, after stringent washing conditions, showed a single species of message of approximately 3 kilobases (data not shown), consistent with previous findings (Vittala et al., 1988). Immunoblot analysis of platelet and endothelial cell lysates (Fig. 2) showed that LAMP-1 in these cells, similar to purified human hepatic LAMP-1, migrated as a broad band of about 110 kDa. Some heterogeneity in the size of the protein from these different cell types was observed and is probably a result of differential glycosylation. There was no reactivity with control antibody at this molecular weight (not shown).

Intracellular Localization and Surface Expression of Platelet

![Fig. 1. Platelet reactivity of the screening IgG before and after affinity purification. Gel-filtered human platelets were treated with thrombin (0.5 unit/ml) or buffer and then incubated with either the rabbit anti-platelet IgG used to screen the cDNA library (panel A) or an IgG fraction affinity purified on the β-galactosidase fusion protein (panel B). After washing and fixation, the platelets were incubated with FITC-conjugated goat anti-rabbit IgG and bound fluorescence analyzed by flow cytometry. Data is expressed as cell number (y axis) as a function of fluorescence on a log scale. The screening IgG (A) bound to the surface of both resting and activated platelets, while the affinity purified antibody (B) bound only to the surface of activated platelets.](http://www.jbc.org/Downloaded from http://www.jbc.org/ by guest on March 24, 2020)
LAMP-1—Murine monoclonal antibody to human LAMP-1 was used to analyze platelet LAMP-1 localization and expression. Flow cytometry studies demonstrated binding to the surface of activated but not resting platelets (Fig. 3A), confirming the results with fusion protein affinity purified polyclonal IgG. Immunofluorescent microscopy of intact and saponin-permeabilized resting platelets showed no binding to intact platelets, but intracellular granular fluorescence of permeabilized platelets (Fig. 3B). These data suggest that LAMP-1 is translocated from an intracellular granular compartment to the surface upon stimulation.

To quantitate LAMP-1 expression, binding of ¹²⁵I-conjugated monoclonal anti-LAMP-1 IgG to activated and resting platelets was measured. Binding was time- and concentration-dependent, and as seen in a representative equilibrium binding isotherm (Fig. 4), reached saturation between 1.5 and 2.0 μg/ml. In this study, nonspecific binding at maximum input antibody concentration was 0.9% (901 cpm). Scatchard analysis (inset) of these data revealed 2190 binding sites/activated platelet. The number of LAMP-1 molecules expressed on activated platelets varied among individual donors as well as from day-to-day with the same donor. In six separate studies we found a mean ± S.D. of 1175 ± 590 (range 525–2190) binding sites/platelet. Resting platelets expressed little or no LAMP-1 on their surfaces. As shown in Fig. 5, when compared to platelets maximally stimulated by thrombin, an average of 5.4% of binding was seen on resting platelets (range 0–90 molecules/platelet, n = 9).

To correlate LAMP-1 surface expression with release of specific platelet granules, expression was compared with specific markers of α-, dense, and lysosomal granules as a function of agonist (thrombin) concentration (Fig. 6). LAMP-1 expression was half-maximal at 0.05 unit/ml thrombin. This clearly differed from both α- and dense granule release. Surface expression of PADGEM (GMP140), a marker for α-granule release, was half-maximal at a 10-fold lower thrombin concentration (0.005 unit/ml), while the half-maximal release of serotonin ([¹⁴C]5-HT), a marker of dense granules occurred at a 5-fold higher thrombin concentration (0.25 unit/ml). In these studies, maximum total binding of ¹²⁵I-anti-PADGEM was 1,100 cpm/aliquot and of ¹²⁵I-anti-LAMP-1 was 11,800 cpm/aliquot. Nonspecific binding was 14 and 4% of the total, respectively. Total [¹⁴C]hydroxytryptamine uptake was 12,100 dpm, nonspecific release was 10%, while maximum specific release was 87%. These data suggest that LAMP-1 translocated from a different compartment than α- or dense granules.

Platelet lysosomal acid hydrolase release has been studied...
by several investigators and found to differ from α- and dense granule release in several respects in addition to thrombin sensitivity. Unlike α- and dense granule secretion, lysosome enzyme release did not follow stimulation by the weak agonists ADP or epinephrine (Kaplan et al., 1979). As shown in Fig. 5, we found that only 10% of maximal thrombin-induced lysosome expression was detected in response to platelet stimulation by ADP or epinephrine. These platelets secreted thrombospondin normally (measured by ELISA) and expressed surface thrombospondin (measured by flow cytometry) indicating that both agonists induced typical activation (data not shown). Also, consistent with published observations of lysosomal enzyme release, we found that LAMP-1 surface expression was partially dependent on the presence of extracellular calcium (Fig. 5) and was insensitive to cyclo-oxygenase inhibition by indomethacin (not shown). Thrombin concentration in the indomethacin studies was low (10–20 milliliters/ml). At these concentrations, α-granule release (measured as TSP secretion) was inhibited by 90%, while

**Fig. 4.** 125I-Anti-LAMP-1 IgG binding to thrombin-stimulated platelets. Gel-filtered platelets were stimulated with 0.5 unit/ml thrombin for 10 min at 22 °C. Increasing concentrations of 125I-murine monoclonal anti-LAMP-1 IgG were then added to 900-μl aliquots for 30 min at 22 °C. Bound radioactivity was then measured after centrifugation through silicone oil. Data are expressed as specific binding, i.e. that which was inhibited by a 20-fold excess of unlabeled antibody. Inset, Scatchard analysis of these data.

**Fig. 5.** Platelet surface LAMP-1 expression. Gel-filtered platelets (300 μl) were incubated with thrombin (0.5 unit/ml), ADP (2.5 μM), epinephrine (0.5 mM), or thrombin (0.5 unit/ml in a calcium-free buffer) for 10 min at 22 °C. 125I-Anti-LAMP-1 IgG was added for 30 min at 22 °C without stirring. Bound radioactivity was then measured after the platelets were centrifuged through silicone oil. Binding is expressed as % maximal specific binding induced by 0.5 unit/ml thrombin in buffer containing 1 mM CaCl2. Specific binding was that inhibited by 20-fold excess of unlabeled antibody.

LAMP-1 surface expression was not inhibited. These data suggest that LAMP-1 translocated from the lysosomal membrane, not α- or dense granule membrane.

A series of sucrose density gradient platelet fractionation studies were done to investigate further the localization of LAMP-1. Platelets were sonicated in such a way as to only disrupt their outer plasma membrane and centrifuged through a 30–60% continuous sucrose gradient. A typical banding pattern was observed. Two narrow, low density “membrane” bands (fractions 2 and 4) were clearly distinguished from an intermediate density band (fraction 6) and two high density bands (fractions 8 and 10). These bands were collected along with the intervening regions (fractions 1, 3, 5, 7, 9, 11–13), and subjected to protein and ELISA analyses. In one experiment, platelets were loaded with [3H]hydroxytryptamine and radioactivity in the fractions detected by scintillation counting. Fig. 7, panel A, demonstrates colocalization of β-galactosidase, a lysosomal enzyme, with LAMP-1, in fraction 6. The M indicates the peak fraction in which GPlb, a marker of the external platelet plasma membrane, was found. Panel B shows the distribution of [3H]hydroxytryptamine, a dense granule marker. Greater than 47% of the total counts recovered were found in one high density fraction, fraction 10. Panel C shows the localization of the α-granule marker, TSP. Greater than 90% of the total TSP was found in fractions 9 and 10. This biochemical pattern of α- and dense granule contents is similar to that reported by others (Van Nostrand et al., 1990). From these data, colocalization of LAMP-1 with a lysosomal enzyme and clear differentiation from the two other granule pools, we conclude that LAMP-1 is lysosomal in platelets.

We have also studied LAMP-1 expression on platelets from a patient with Hermansky-Pudlak syndrome, an inherited platelet storage pool disease. Platelets from these patients have been described as having diminished or absent ability to secrete dense granule and lysosomal contents (Rendu et al., 1987). As shown in Fig. 8, platelets from this patient expressed the α-granule marker PADGEM normally upon thrombin stimulation, but had diminished expression of surface LAMP-1. Immunofluorescence microscopic studies on permeabilized Hermansky-Pudlak resting platelets showed punctate intracellular fluorescence identical to normal platelets, and Western blot analysis of lysates of these platelets with a polyclonal anti-LAMP-1 IgG gave characteristic reactivity (data not
FIG. 7. Sucrose density gradient centrifugation of sonicated platelets. Panel A shows colocalization of β-galactosidase, a lysosomal enzyme, with LAMP-1. The M refers to the peak fraction in which GPⅠb, a plasma membrane marker, was detected. Proteins were detected by ELISA, with specific antibodies, followed by an alkaline phosphatase-conjugated second antibody. The data are expressed as rate of color development (a function of bound antibody) over time. Panel B is the distribution of [3H]hydroxytryptamine (serotonin), a dense granule marker, as measured by scintillation counting. Panel C is the distribution of TSP, an α-granule marker, measured by ELISA, as in panel A.

shown). These data further support the conclusion that LAMP-1 is a platelet lysosomal membrane protein.

DISCUSSION

Using a novel molecular approach, we have identified and cloned a platelet protein expressed on the cell surface only after activation. This 110–120 kDa protein is identical to a previously identified lysosomal membrane protein, LAMP-1. Our data suggest that in platelets this protein is also lysosomal and becomes incorporated into the external plasma membrane during secretion. These data include immunofluorescence studies localizing the antigen to an interior granular compartment in permeabilized, unstimulated platelets and several sets of studies correlating surface expression of LAMP-1 best with lysosomal secretion, not with secretion of either of the other two platelet granular compartments (α- and dense).

Previous studies (Kaplan et al., 1979) have shown that platelet lysosomal acid hydrolase release was clearly discernible from α- and dense granule release by virtue of 1) different dose-response to thrombin stimulation, 2) insensitivity to stimulation by the weak agonists ADP and epinephrine, 3) partial sensitivity to the presence of extracellular calcium, and 4) insensitivity to cyclo-oxygenase inhibition when stimulated by low concentrations of thrombin. Our studies have shown that, similar to platelet lysosomal acid hydrolase secretion, LAMP-1 surface expression was maximal at thrombin concentrations intermediate between those necessary to obtain maximal release of α- or dense granules (Fig. 4). LAMP-1 expression was increased by ~40% when platelets were stimulated in the presence of 1 mM CaCl₂ and was only minimally induced by ADP or epinephrine (Fig. 5). In addition, we have shown colocalization of LAMP-1 with a lysosomal enzyme, β-galactosidase (Fig. 7), after fractionation and sucrose density centrifugation. In this study, there was clear separation of LAMP-1-containing granules from α- and dense granules. Finally, we demonstrated impaired expression of LAMP-1 in Hermansky-Pudlak platelets, which are defective in lysosomal and dense granule release, but showed normal expression of the α-granule membrane protein PADGEM (GMP140).

Secretion in response to a stimulus from cellular storage pools is a general characteristic of many cells, including platelets. In his widely accepted membrane flow hypothesis, Palade suggested that during secretion, granule integral membrane proteins are incorporated into the plasma membrane. PADGEM (GMP140), a platelet α-granule integral membrane protein, provided evidence that a membrane fusion event does indeed occur during platelet activation and secretion (Berman et al., 1986). We report here, for the first time, that LAMP-1,
an integral lysosomal membrane protein, also translocates to the plasma membrane during an activation and secretion event, suggesting that lysosomal membrane fusion also occurs during platelet secretion. Nieuwenhuis et al. (1987) have recently characterized a monoclonal anti-platelet antibody (designated 2.28) that reacts with activated platelets and immunolocalizes to lysosomes. The 53-kDa antigen recognized by this antibody has not yet been molecularly characterized but is thought to be a secreted protein, not an integral membrane protein.

LAMP-1 is one of a family of lysosomal membrane proteins that have been identified and characterized in several species. These include rat lgp110 and 120 (Himeno et al., 1985; Howe et al., 1985), chicken LEPF (Fambrough, 1986), and mouse (Chen et al., 1985) and human LAMP 1 and 2 (Fukuda et al., 1988; Viitala et al., 1988). All are type 1 membrane proteins of 380-396 amino acids with short (10-11 amino acids) cysteic tails. They are heavily N-glycosylated on the intralysosomal domains, with the glycan coat more than half the total protein weight. The proteins have two homologous intralysosomal domains, each with 4 conserved cysteines that form two 36-38 amino acid loops (Fukuda et al., 1988). These two repeated domains are separated by a 25-30 amino acid segment rich in proline and serine or threonine, homologous to the IgA hinge region (Viitala et al., 1988). Granger et al. (1988) have grouped these proteins into two categories: IgA and B, corresponding to human LAMP-1 and -2, respectively. Intraspecies homology within each group is high (e.g. human and mouse LAMP-1 share ~66% homology). Sequence homologies between the two groups, however, are less (human LAMP-1 is ~35% homologous to LAMP-2). These differences are felt to have functional importance, since lysosomes contain proteins from both groups. Recently, it has been shown that LAMP-1 and LAMP-2 are encoded by genes localized to different chromosomes. LAMP-1 was found on chromosome 13q34, and LAMP-2 on Xq24-25. A putative pseudogene for LAMP-1 was also identified on chromosome 12p13 (Mattei et al., 1990). These results, in addition to the rather low sequence homology between LAMP-1 and LAMP-2, support the hypothesis that these two genes diverged early in evolution, and probably have distinct functions. There has been speculation that the heavy glycosylation protects the polypeptide backbone of these proteins, and thus, the lysosomal membrane from hydrolytic enzymes (Carlsson et al., 1988).

While there is no known function for any of these proteins, the amino acid sequence of LAMP-1 is homologous to two leukemia cell glycoproteins, GP130/2B from the highly metastatic tumor cell line MDAY-D2 (Chen et al., 1985) and a leukemia cell glycoprotein, GP130/P2B from the highly metastatic tumor cell line MDAY-DX (Chen et al., 1985) and a leukemia cell line, LS-148, which expresses LAMP-1, that are preferentially expressed on activated cells and may aid in the design of novel diagnostic and therapeutic approaches to diseases such as atherosclerosis and thrombosis.

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