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Pathogenic Cascades in Lysosomal Disease – Why so Complex?

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Summary

Lysosomal disease represents a large group of more than 50 clinically recognized conditions resulting from inborn errors of metabolism affecting the organelle known as the lysosome.The lysosome is an integral part of the larger endosomal/lysosomal system, and is closely allied with the ubiquitin-proteosomal and autophagosomal systems, which together comprise essential cell machinery for substrate degradation and recycling, homeostatic control, as well as signaling. More than two-thirds of lysosomal diseases affect the brain, with neurons appearing particularly vulnerable to lysosomal compromise and showing diverse consequences ranging from specific axonal and dendritic abnormalities to neuron death. While failure of lysosomal function characteristically leads to lysosomal storage, new studies argue that lysosomal diseases may also be appropriately viewed as "states of deficiency" rather than simply overabundance (storage). Interference with signaling events and salvage processing normally controlled by the endosomal/lysosomal system may represent key mechanisms accounting for the inherent complexity of lysosomal disorders. Analysis of lysosomal disease pathogenesis provides a unique window through which to observe the importance of the greater lysosomal system for normal cell health.

Introduction

Lysosomal disease encompasses a wide spectrum of rare genetic disorders with defects in proteins essential for normal function of the lysosomal system (Platt and Walkley, 2004). Most lysosomal diseases show widespread tissue and organ involvement, with brain, viscera, bone and connective tissues often being affected. Brain disease is particularly prevalent, involving two-thirds of all lysosomal diseases. Here, clinical disease may be manifest as mental retardation and/or dementia, sensory loss including blindness or deafness, motor system dysfunction, seizures, sleep and behavioral disturbances, and so forth. Lysosomal diseases are progressive, and ultimately fatal, disorders but affected individuals generally appear normal at birth and develop clinical disease only years or decades later. The disease course can be highly variable even among affected siblings in the same family, and more often than not clear-cut genotype-phenotype correlations have not been established. That such multifaceted and serious disease conditions result from simple inborn errors of metabolism raises the persisting question of why these disorders are ultimately so clinically complex. A predominate early view to explain lysosomal disease pathogenesis emanated from the original concept that these disorders are caused by individual lysosomal enzyme deficiencies followed by accumulation of a single major substrate normally degraded by that enzyme (Hers, 1965). Following the build-up of this non-degraded primary substrate, the endosomal/lysosomal (E/L) system would eventually be overwhelmed, normal cell functions would collapse, and cells would simply die as a result of this progressive constipation. This so-called 'Cytotoxicity Hypothesis" (Desnick, et al., 1976) is still often cited today to explain the pathogenesis of lysosomal disease, yet modern developments in cell biology and neuroscience offer much richer avenues of explanation. For lysosomal diseases affecting brain, for example, four central themes can be recognized as setting the stage for this complexity in pathogenic cascades. These are based on: (i) the wide

variety of enzyme and non-enzyme proteins implicated in causing lysosomal disease and the many ways defects in their expression or function can compromise the lysosomal system, (ii) the complexity of the brain itself in terms of heterogeneity of neuronal and glial cell types which typically exhibit distinct metabolic identities and interrelationships, and (iii–iv) the potential roles of the greater E/L system in signal transduction and in cellular homeostatic control, and the consequences following compromise in these functions in lysosomal disease.

Defective proteins causing lysosomal disease are widely varied in function and location

The range of molecular defects causing lysosomal compromise in lysosomal disease is extensive and involves both soluble and membrane-associated proteins (Platt and Walkley, 2004). Indeed, defects in no less than 50 different proteins have been implicated to date as causing lysosomal dysfunction, and new proteins linked to lysosomal disease continue to be identified. While most are enzymes with acidic pH optima consistent with the original lysosomal disease concept, many others are not. Included here, for example, are enzymes in the Golgi-TGN or ER that are responsible for trafficking and catalytically activating specific lysosomal enzymes. Defects in these proteins cause mucolipidosis (ML) II/III and multiple sulfatase deficiency (MSD), respectively. Lysosomal diseases are also caused by defects in soluble and membrane-associated non-enzyme proteins of late endosomes and lysosomes believed essential for the processes of substrate degradation and egress, as well as vesicle fusion. Examples of soluble proteins are prosaponin and its processed components, saposins A–D, and the GM2 activator protein (GM2-AP), all of which are critical for aiding in the enzymatic degradation of glycolipid substrates. The soluble protein, NPC2, is believed critical for cholesterol egress, but so too is the membrane-associated NPC1 protein, as discussed below.

The complexity of the molecular underpinnings of lysosomal disease is typified by conditions like GM2 gangliosidosis and Niemann-Pick diseases. GM2 gangliosidosis can be caused by defects in β-hexosaminidase (composed of two subunits, α and β, coded for by two different genes) or by defects in the GM2-AP. Hence the occurrence of Tay-Sachs (α subunit defect), Sandhoff (β-subunit defect) or the AB-variant (GM2-AP defect) diseases, all of which nonetheless manifest as GM2 gangliosidosis. Niemann-Pick diseases are similarly complex (Walkley and Suzuki, 2004). Type C disease, originally named due to its clinical similarity to Niemann-Pick A/B disease, was later found not to exhibit a primary defect in sphingomyelinase as occurs in A/B disease, but rather was due to defects in the aforementioned cholesterol binding proteins (NPC1 or NPC2) of the late endosome. Interestingly, the NPC1 and 2 proteins are dramatically different, one being a transmembrane protein (NPC1) of late endosomes, the other a soluble protein (NPC2) in the lumen of late endosomes. When either protein is defective the same apparent cholesterol storage condition ensues, one believed caused by a block in retroendocytic trafficking of cholesterol out of late endosomes to other sites in the cell (Pentchev, et al., 1994). Cholesterol storage is accompanied by accumulation of both acidic and neutral glycosphingolipids (GM2 and GM3 gangliosides; also lactosylceramide). And while unesterified cholesterol is typically viewed as the primary storage material in Niemann-Pick C disease, experiments limiting expression of complex gangliosides lead to significant reductions in the extent of cholesterol sequestration in most neurons (Gondré-Lewis and Walkley, 2003). This suggests that the NPC1 and NPC2 proteins may also be associated with retroendocytic trafficking of GSLs. Interestingly, cholesterol sequestration in neurons is also prominent in the GM1 and GM2 gangliosidoses, but here clearly occurs secondary to the primary storage of gangliosides.

Like NPC1 and NPC2 described above, defects in other enigmatic proteins have similarly been linked to lysosomal disease. For example, mutations in the *MCOLN1* gene which codes mucolipin-1, a lysosomal membrane TRP (Transient Receptor Potential) family of ion channel,

causes mucolipidosis type IV (MLIV) disease (Zeevi, et al., 2007). In spite of its name MLIV has little connection other than historic with MLII/III diseases which are caused by defects in the phosphotransferase enzyme responsible for adding the mannose-6 phosphate moiety to lysosomal enzymes as required for normal targeting to lysosomes. Like NPC1, mucolipin-1 resides in the membrane of late endosomes/lysosomes and while implicated in lysosomal pH control and in membrane fusion/fission events, its function remains essentially unknown. A similar situation exists for many of the proteins implicated in the ten (CLN1-CLN10) so-called neuronal ceroid lipofuscinoses, or Batten diseases (Kyttälä, et al., 2006). The CLN3 protein, for example, defects in which cause juvenile Batten disease, may be associated with autophagolysosomes fusion/maturation (Cao, et al., 2006), in lysosomal pH control (Pearce, et al., 1999), or a host of other functions (Rakheja, et al., 2008). Similarly, the CLN6 and CLN8 proteins are believed localized to membranes of the ER and while their absence leads to lysosomal storage, their functional link to lysosomes is unknown (Kyttälä, et al., 2006).

Thus it is evident from the above, brief overview that defective proteins in lysosomal diseases vary widely and the consequences of their functional loss are often not understood (Platt and Walkley, 2004). While identification of primary substrates has provided insight into identification of lysosomal enzyme defects, when lysosomal diseases are caused by proteins responsible for other functions – like lysosomal pH control (as an example) – primary substrates may be heterogeneous. Yet even for lysosomal hydrolase defects the materials accumulating in the E/L system are also remarkably heterogeneous. Numerous gangliosides, for example, accumulate in neurons in most of the mucopolysaccharidoses (MPSs) even though the enzymes defective in these conditions are known to act on glycosaminoglycan (GAG) degradation and not GSLs (Neufeld and Muenzer, 2001). Secondary inhibition of GSL degradative enzymes by the primary GAG storage has been hypothesized, but the sequestered gangliosides (GM2 and GM3) do not typically localize with each other, or with GAG storage (McGlynn et al., 2004). Unesterified cholesterol also accompanies this storage in MPS disease, possibly secondary to the gangliosides and in a manner similar to what occurs in GM2 gangliosidosis described above. Remarkably, ganglioside and cholesterol storage also occurs in brain in the glycoproteinosis known as α-mannosidosis, again without explanation (Walkley, 2004). Secondary storage of glycolipids and related materials is actually the rule rather than the exception in lysosomal disease, and likely contributes significantly to compromise in cell function, as described below.

Lysosomal diseases often exhibit selective neuronal and glial involvement

While some lysosomal diseases, like Niemann-Pick type C, exhibit storage and related consequences in cells throughout the brain, other diseases exhibit a much more restricted distribution. Examples of this selective vulnerability to lysosomal dysfunction abound. Fabry disease, for example, in terms of CNS involvement, has long been known to show storage of its primary substrate (globotriaosylceramide) largely limited to scattered neurons in the spinal cord, brainstem, amygdala, hypothalamus and entorhinal cortex (deVeber, et al., 1992). In neuropathic Gaucher disease storage in neurons is minimal to absent but storage in cells of monocyte/macrophage lineage, the so-called Gaucher cells, is prominent (Beutler and Grabowski, 2001). In Krabbe disease, like Gaucher, neuronal storage is not evident and the presence of swollen macrophages (globoid cells) predominates (Wenger, et al., 2001). In addition there is widespread death of oligodendrolia, possibly secondary to the buildup of the toxic compound, psychosine, leading to myelin deficits (demyelination) and in resulting clinical disease. Most lysosomal diseases also exhibit activation of microglia and the debate is ongoing as to whether this activation is caused by metabolic events intrinsic to microglia (and therefore inappropriate) or, alternatively, is in direct and appropriate response to abnormalities in neurons (e.g., altered surface expression of gangliosides, apoptotic or necrotic death, etc.). Understanding events triggering inflammation is of major importance since therapies directed

at ameliorating microglial activation presumably could be beneficial in some circumstances but possibly detrimental in others. Such events may also vary as lysosomal diseases progress.

In some lysosomal diseases, primary storage in neurons is widespread but restricted cell populations selectively exhibit secondary accumulation of additional compounds. For example, in the glycoproteinosis, α-mannosidosis, mentioned earlier, storage of characteristic mannose-rich oligosaccharides is widespread and affects all cells but secondary storage of GSLs is a conspicuous feature of select subpopulations of cortical pyramidal neurons and some GABAergic intrinsic neurons (Walkley, 2004). These ganglioside-sequestering cortical pyramidal neurons have been shown to sprout ectopic dendrites in a manner similar to what happens to essentially all cortical pyramidal neurons in primary GM1 and GM2 gangliosidosis. This growth of ectopic dendrites, a phenomenon unique to lysosomal disease, is always found limited to GSL-storing cortical pyramidal neurons and multipolar neurons in the claustrum and amygdala (Walkley, et al, 2000; Walkley, 2003; Walkley, 2004).

Just as cortical pyramidal neurons selectively show the presence of renewed dendrite growth in some lysosomal diseases, other neurons exhibit unusual axonal alterations known as neuroaxonal dystrophy or axonal spheroid formation (Walkley, et al., 1991; Walkley, 2004). Spheroid formation appears detrimental to neurons, since Purkinje cells, which appear particularly vulnerable to spheroid formation, also tend to die early in the disease process. Ultrastructural studies have revealed that spheroids are not composed of materials like those accumulating in perikarya (membranous cytoplasmic bodies, zebra bodies, etc.), but rather consist of collections of tubulovesicular profiles, mitochondria, and autophagosomal-like and multivesicular-type bodies. This ultrastructure is similar from one storage disease to the next consistent with a generic cause for their formation (Walkley, 2004). The incidence and distribution of spheroids correlate closely with the onset and type of motor system defects exhibited by affected animals, suggesting that they are a major player in generating neurological dysfunction. Spheroids are readily visualized in neurons using immunocytochemistry to label components trafficked in axons, including enzymes (glutamic acid decarboxylase) as well as calcium binding proteins (parvalbumin, calbindin). Using these cell selective markers, spheroids were found to occur most commonly on GABAergic neurons, including Purkinje cells, neurons in basal ganglia, and nonpyramidal (intrinsic) cells of cerebral cortex. Spheroids are well documented in lysosomal disorders where they are found in abundance in primary ganglioside storage diseases, Niemann-Pick disorders (types A and C) and in the glycoproteinosis, α-mannosidosis. In other lysosomal diseases, most notably the early-onset neuronal ceroid lipofuscinoses (CLN1 and CLN2 diseases), spheroid formation is not characteristic but widespread neuron death in cerebral cortex is a common occurrence. Thus whether the result of the primary metabolic event, or a downstream consequence in the disease cascade, selective neuronal vulnerability in lysosomal disease is a characteristic finding. This feature of lysosomal disease undoubtedly reflects significant differences in the metabolic signatures of individual types of neurons comprising the complex neuronal networks of the brain. Eventually it should be possible to use the presence of these specific disease features to gain insight into the specialized characteristics of normal neurons. Ectopic dendritogenesis, for example, suggests the presence of a unique capacity for dendritic plasticity in the affected pyramidal cell populations in cerebral cortex, amygdala and claustrum. Understanding what is being triggered in lysosomal disease to cause a renewal of dendritic sprouting and associated synapse formation may provide novel insight into broader mechanisms underlying learning and memory in normal brain.

The role of the E/L system in signal transduction events

Not only has our understanding of the complexity of lysosomal diseases and their defects expanded significantly in recent years, but so too has knowledge of the role of the E/L system

in overall cell function (Pol and Di Fiore, 2006) (Fig. 1). In addition to being involved in regulating the composition of neuronal membranes and in internalizing receptor-ligand complexes, endocytosis also plays a critical role in attenuating and integrating a wide variety of signaling events affecting many cellular functions. In neurons, endocytic events similarly govern a variety of signaling mechanisms, with some like those associated with neurotransmitter actions being unique to these types of cells. For example, endocytic mechanisms are known to control the availability of neurotransmitter receptors at excitatory synapses. AMPA (α-amino-3-hydroxy-5-methylisoazole-4-proprionic acid) receptors, which regulate the majority of fast excitatory neurotransmission and are therefore critical for synaptic plasticity, are known to undergo rapid constitutive internalization as a consequence of synaptic activity. Once internalized, AMPA receptors are sorted from early endosomes to either specialized recycling endosomes for re-insertion in the plasmalemma or, are trafficked to late endosomes for fusion with lysosomes and resulting elimination (Barry and Ziff, 2002; Bredt and Nicoll, 2003). In contrast, the constitutive internalization of NMDA (N-methyl-d-aspartate) and metabotropic glutamate receptors (mGluRs) is less prominent. Subunits forming AMPARs (GluR1-4) are well documented to assemble as homo- or hetero-dimers, with subunit composition being critical for their functional properties. The GluR2 subunit, unlike GluR1, 3 and 4, is Ca^{2+}-impermeable, making it the key determinant of AMPAR function (Isaac, et al., 2007). Importantly, over-expression of the GluR2 subunit in cultured neurons induces ectopic dendritic spine formation, consistent with AMPA receptor expression and localization playing a major role in dendrite and synapse plasticity (Passafaro, et al., 2003). These findings suggest that the E/L system – to the extent that it influences AMPA receptor availability - is poised to exert significant influence over dendritic plasticity. Given that many lysosomal diseases are characterized by ectopic dendritogenesis raises important questions about possible compromise in AMPA receptor cycling secondary to lysosomal system compromise (Walkley, 2007).

In addition to a possible compromise in neurotransmitter receptor recycling, another prominent aspect of signaling that may be altered in lysosomal disease involves growth factors. Growth factor receptors are known to be internalized by endocytosis, with this occurring both in the soma-dendritic as well as axonal domains, with signaling events varying according to the specifics of the endocytic route and vesicle type (Bronfman et al., 2006). Importantly, studies have shown that growth factors like BDNF, a TrkB ligand, when applied to young cortical neurons can cause exuberant dendrite growth (McAllister, 2001). As in the example of AMPA receptor trafficking and the E/L system described above, this observation raises the question of whether growth factor signaling might be abnormal in lysosomal disease as a way to account for the abnormal growth of dendrites. Growth factors are also endocytosed at synaptic terminals and transported along with their receptors back to the neuronal cell body for signaling (Bonfman, et al., 2006). These retrogradely transported vesicles are often referred to as signaling endosomes since they contain ligand-bound growth factor receptors derived from postsynaptic sites which are being transported back to the cell body where they are believed to recruit appropriate second messenger cascades for signal transduction purposes (Howe and Mobley, 2005). The prominence of spheroids in axons of Purkinje cells, described earlier, and the finding that these cells are prone to die early in many storage diseases have suggested that the spheroids themselves may be contributing to this early demise by causing a block in signaling endosomes carrying a growth factor/receptor complex essential for cell survival (Walkley, 2004). Thus signaling events in lysosomal disease may be exaggerated, as suggested by the occurrence of ectopic dendritogenesis, or compromised, as suggested by Purkinje cell death following spheroid formation. Clearly these features of lysosomal disease pathogenesis deserve further investigation.

The "Greater Lysosomal System" as a central metabolic coordinator

It can be argued that the primary goal of lysosomal processing is the degradation and recycling of breakdown products for biosynthetic purposes, a process aptly referred to as "metabolic salvage" (Tettamanti, et al., 2003) (Fig. 1). For some of these byproducts of lysosomal catabolism, e.g., free sialic acid and cystine, specific transport proteins have been identified (sialin and cystinosin, respectively) whose importance is made apparent by diseases caused by their absence (Platt and Walkley, 2004). Yet, there is ample evidence that some complex molecules processed by the E/L system do not have to be degraded to their simplest components prior to exit from these organelles. Prime examples are gangliosides which are synthesized in the Golgi-TGN and delivered to the cell surface by vesicular transport in exocytic vacuoles, followed by insertion in the outer leaflet of the plasmalemma where they reside in close association with membrane raft-related molecules, including cholesterol, sphingomyelin, and various GPI-anchored proteins (Schwarzmann and Sandhoff, 1990; Kolter and Sandhoff, 2005). Following endocytosis, complex gangliosides come into contact with an array of hydrolytic enzymes and activator proteins that facilitate their catabolism to monosialogangliosides (e.g., GM1). Subsequently, GM1 is degraded to GM2 by lysosomal β-galactosidase and GM2 to GM3 by lysosomal β-hexosamindase (in conjunction with the GM2-AP), with defects at either of these degradative steps leading to GM1 and GM2 gangliosidosis, respectively. Numerous studies on the fate of less complex gangliosides (GM2 and GM3, as well as GM1) and of additional breakdown products of gangliosides (neutral glycosphingolipids known as lactosylceramide and glucosylceramide) indicate that they may be recycled (or "salvaged") after endocytosis prior to their complete degradation (Tettamanti, et al., 2003). Thus, gangliosides like GM2 and GM3 may exit the E/L system and be trafficked to the Golgi/TGN where they would be reglycosylated and delivered again to the plasmalemma. Importantly, this suggests that there are two potential routes available to monosialogangliosides as they transit through the lysosomal system - one involving direct recycling to the Golgi/TGN and the other leading to full lysosomal degradation.

Given the above, an emerging question of central importance for lysosomal disease is as follows: What are the consequences of a failure to recycle material (e.g., monosialogangliosides, cholesterol, GAGs, other) out of the E/L system? This question would be easier to answer if there was greater understanding of the function of gangliosides in neurons, or if the relationship between GAG recycling and proteoglycan synthesis and function were clearly defined, or if the routes of trafficking of unesterified cholesterol within neurons or between neurons and astrocytes was fully known. But even with these significant limitations, several scenarios concerning this apparent 'failure to recycle' can be considered. The first of these, as stated earlier, is that lysosomal storage simply overwhelms the cell's capacity for volume expansion and causes death as a consequence. A second possibility gaining strength with recent publications is that sequestered substrates like GM1 ganglioside in GM1 gangliosidosis may "leak" into membrane domains normally containing little or no ganglioside, like the ER, resulting in depletion of Ca^{++} stores, activation of the ER stress response and eventually apoptosis and neuron death (d'Azzo, et al., 2006). Similarly, in GM2 gangliosidosis it has been shown that GM2 increases in microsomal membranes inhibit the activity of SERCA, which could similarly cause ER stress and apoptosis (Ginzburg, et al., 2004). Parallel events in dysregulation of intracellular stores of Ca^{++} secondary to inappropriate ganglioside build-up in internal cell membranes may similarly be occurring in Gaucher, Niemann-Pick A and other lysosomal diseases. Changes in ganglioside expression or availability at the plasmalemma may also be occurring, followed by alteration in surface receptor expression (e.g., of toll-like receptors) and microglial activation (Jou, et al., 2006).

While the above events may prove of significant impact in lysosomal disease, these conditions nonetheless are typically chronic and most neurons exist for many years in the face of slowly

progressive lysosomal storage. Thus, to a great extent, pathogenic cascade events that are occurring are not acutely cytotoxic. They also likely are highly varied across the spectrum of lysosomal disease. For example, recent studies show that the brains of mice lacking the NPC1 protein and developing cholesterol/GSL storage typical of Niemann-Pick C disease are deficient in the neurosteroid, allopregnanolone (Griffin, et al., 2004). Since allopregnanolone is a neurosteroid requiring cholesterol for synthesis, this finding has been interpreted to suggest that the block in cholesterol movement out of late endosomes to mitochondria (where the synthetic enzymes for allopregnanolone production are found) is responsible for this deficit. Such findings have also suggested a possible new form of therapy based on byproduct replacement, in which missing compounds are administered (Griffin, et al., 3004; Walkley, 2007). Allopregnanolone may be but one example of a metabolic product deficit occurring secondary to lysosomal storage. Indeed, GSLs may represent a second class of compounds impacted by sequestration of metabolic precursors, as suggested many years ago by Sandhoff and colleagues (Schwarzmann and Sandhoff, 1990). As it has been estimated that as much as 70% of the GSL pool in neurons is salvaged prior to complete degradation in the E/L system, the sequestration of simple gangliosides (GM1, GM2, GM3) in lysosomal disease may result in a deficit of precursor material in the Golgi/TGN for renewal of complex gangliosides at the plasmalemma. One consequence of such an event might be for the neuron to up-regulate GSL synthesis, an event that could in turn alter the expression of specific gangliosides or related components of membrane rafts in the plasmalemma. Enhanced GSL synthesis, if it occurred, likely would not solve the neuron's deficit, however, but rather would simply add further to the storage process.

Yet another solution to metabolic precursor deficits might be for the neuron to "mine" resources within the cell through a mechanism known as macroautophagy (autophagy) (Fig. 1). Recent studies have shown that maintaining a basal level of autophagy is critical for normal neuronal function (Hara, et al., 2006;Komatsu, et al., 2006), so in this respect they are not really different from other cells in which starvation-induced stress elicits autophagy (Cuervo, 2004). Macroautophagy is also closely allied to protein degradative mechanisms associated with Chaperone-Mediated Autophagy (CMA) and the Ubiquitin-Proteosomal System (UPS). The involvement of autophagy, as well as possible CMA and UPS defects, has recently come under increased focus in lysosomal disease (Pacheco and Lieberman, 2007;Pacheco, et al., 2007;Settembre et al., 2008;Tayebi, et al., 2003). In terms of autophagy, current reports support the possibility that autophagy is blocked as a general feature of lysosomal storage or, alternatively, is increased. Blockage in autophagy clearly would be detrimental to neurons, but similarly, if autophagy is increased, this again may not be beneficial since ultimately the same catabolic defect, in the case of lysosomal enzyme deficiencies, would limit access to the stored GSLs. In the case of Niemann-Pick C disease, or other putative substrate trafficking-type defects, however, autophagy might offer an alternative route for allowing interaction between sequestered GSLs and normal catabolic enzymes present in lysosomes. That is, autophagy could, conceivably, be playing a role in partially ameliorating storage in some lysosomal diseases. If so, pharmacological enhancement of autophagy might provide additional benefit, as recently suggested for Huntington disease (Ravikumar and Rubinstein, 2006).

Whether the neuron is increasing activity in synthetic pathways or enhancing autophagy in an effort to overcome lysosomal storage, such actions would be expected to be energy consuming. Interestingly, recent studies have demonstrated that mouse models of lysosomal disease exhibit reduced fat stores unrelated to compromise in food intake, consistent with the presence of an energy consuming process inherent in these diseases (Woloszynek, et al., 2007). As a whole these studies illustrate the array of events that may contribute to the complex disease cascades in lysosomal disease and raise the interesting question of whether these disorders are more appropriately viewed as disease-induced states of "starvation stress" – that is, as conditions characterized by deficiency, rather than overabundance (storage).

Conclusions

Growth of knowledge about lysosomal disease mirrors the modern advance of Medicine begun by Garrod with the discovery of inborn errors of metabolism (Garrod, 1909). Over the past century there have been eras focused on clinical descriptions, on biochemical characterization of enzyme defects and primary storage materials and, most recently, on gene and mutation analysis. Now, it can be declared, we have entered a new era, one that draws on all of these earlier discoveries and begins to address, in detail, the disordered events in cell biology brought about by individual lysosomal system defects – *the era of lysosomal disease pathogenesis*. Clearly, a wide array of protein defects leads to metabolic compromise and storage in lysosomal disease, with these events in turn driving the development of specific cellular pathologies depending on the cells affected and their metabolic signatures. Disruption of E/L function in neurons has the capacity to compromise many normal cell operations and to generate a host of downstream consequences from the growth of ectopic dendrites, to formation of axonal spheroids, to neuron death, and so forth. Failure to recycle materials out of the E/L system in lysosomal disease may also lead to deficiency of precursor pools of metabolites and thereby alter cellular homeostatic functions leading to changes in synthetic pathways and/or to autophagy in an effort to overcome these deficits. Analysis of these complex pathogenic cascades in lysosomal disease will offer new insights into therapy as well as the opportunity to more fully appreciate the role of the greater lysosomal system in cells of the normal nervous system.

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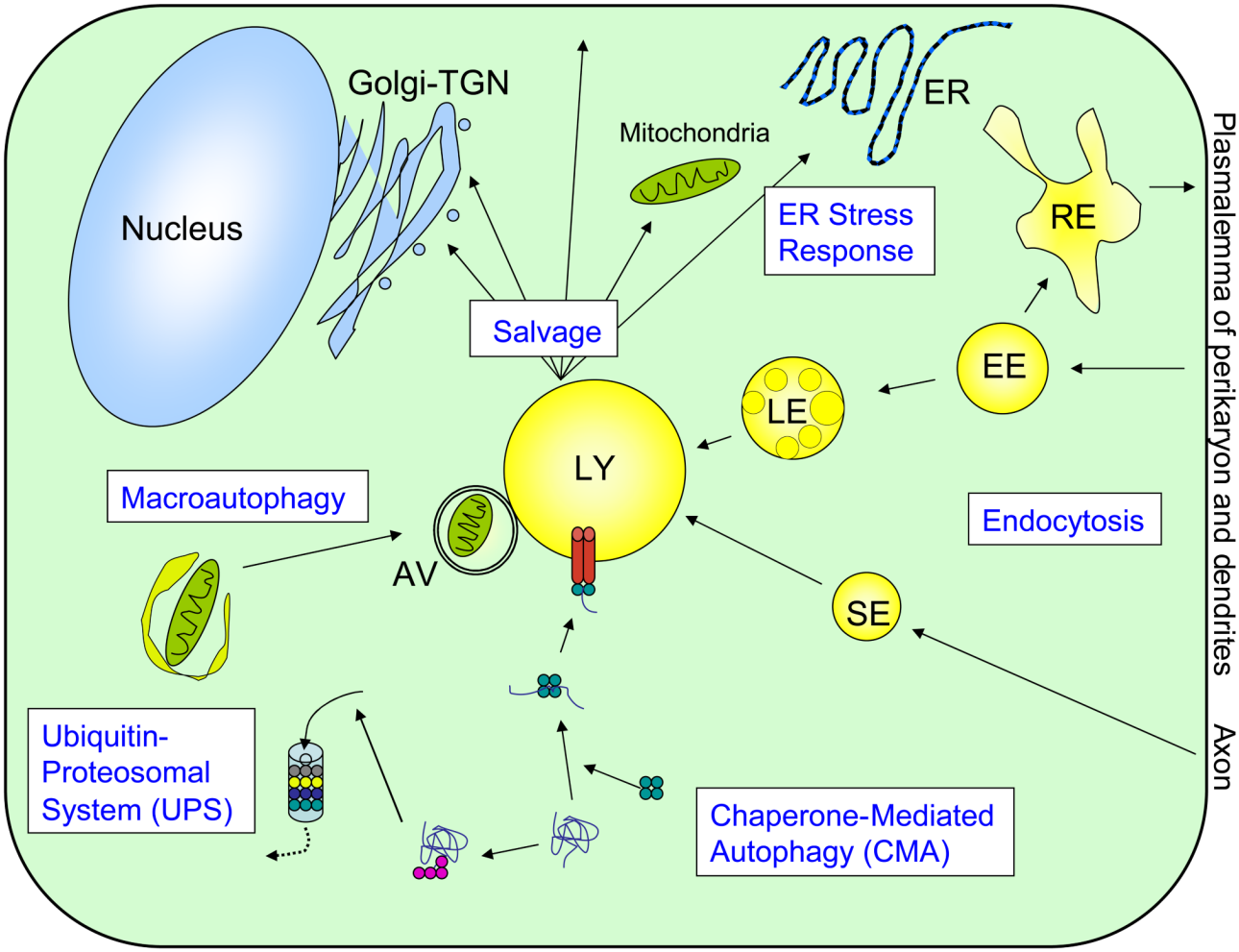

Figure 1.
Schematic summarizing the concept of the lysosome as a central element in the greater lysosomal system of neurons with links to the endocytic streams from somadendritic and axonal domains, and to the macroautophagy stream and its close allies, the ubiquitin-proteosomal system (UPS) and chaperone-mediated autophagy (CMA) components. What flows into this system must also leave in some form, depicted here as the salvage pathway with delivery to the Golgi/TGN, mitochondria, and other sites in the cell. The complexity of the disease cascades in lysosomal disorders is conjectured to emanate in part from disruption of these interrelated components of the greater lysosomal system, as described in the text. (EE, early endosome; RE, recycling endosome; LE, late endosome; LY, lysosome; SE, signaling endosome; AV, autophagic vacuole; ER, endoplasmic reticulum).