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Review

Membrane Traffic and Muscle: Lessons from Human Disease

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Like all mammalian tissues, skeletal muscle is dependent on membrane traffic for proper development and homeostasis. This fact is underscored by the observation that several human diseases of the skeletal muscle are caused by mutations in gene products of the membrane trafficking machinery. An examination of these diseases and the proteins that underlie them is instructive both in terms of determining disease pathogenesis and of understanding the normal aspects of muscle biology regulated by membrane traffic. This review highlights our current understanding of the trafficking genes responsible for human myopathies.

Key words: BIN1, caveolin-3, dynamin 2, dysferlin, membrane traffic, myotubularin

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Skeletal muscle is a highly specialized tissue with a complex structure designed for the generation of force. Like all organ systems, skeletal muscle uses the basal elements of membrane traffic, including forward transport of newly synthesized proteins, internalization of receptors and growth factors, and processing of proteins and membranes for degradation via the lysosome (1). However, because of its unusual structure, with individual myofibers containing multiple nuclei and the majority of the cytoplasm dedicated to the contractile apparatus, muscle is a unique setting in which this trafficking machinery operates. The Golgi apparatus, the endoplasmic reticulum and the endosomes are in tightly restricted regions of the myofiber, and require extensive organelle rearrangement during myogenesis to arrive at their location in the mature cell (2). In addition, there are several processes specific to muscle that require membrane traffic. These include the establishment and maintenance of the neuromuscular junction (3), the formation and function of the T-tubule (4), and the generation and repair of myotubes via membrane and myoblast fusion (5).

The importance of membrane traffic for muscle is highlighted by the fact that several muscle diseases are the result of defects in its machinery. These disorders illuminate some of the specific roles that membrane traffic plays in the development and maintenance of the myofiber. Conversely, in certain cases they bring out the disparity between our general knowledge of trafficking processes and our specific understanding of the pathogenic mechanisms that underlie these diseases. The purpose of this review was to discuss the muscle disorders caused by mutations in the trafficking machinery and to describe the theories related to the functions and abnormalities of the individual disease-associated gene products. We will consider the dysferlinopathies (Table 1) (6), the caveolinopathies (Table 2) (7), and a family of diseases called centronuclear myopathies (Figure 1), all of which result from mutations in several trafficking genes (8).

Dysferlin

Dysferlinopathies

The dysferlinopathies are a spectrum of autosomal recessive muscle diseases caused by mutations in the dysferlin gene (6). The primary dysferlinopathies are limb girdle muscular dystrophy (LGMD) type 2B (LGMD2B) and Miyoshi myopathy (MM) (9,10). LGMD2B is a disorder characterized by progressive proximal muscle weakness and atrophy (proximal muscles = shoulder, upper arm, pelvic and thigh muscles), with onset in adolescence or early adulthood. MM, however, begins with weakness and atrophy in the distal posterior muscles (especially the calf muscles), with spread to the proximal musculature late in the condition. While the two diseases are clearly distinct clinical entities, they share some features: extremely high elevations of creatine kinase (a serum marker for muscle breakdown), similar age of onset (mid/late childhood to early adulthood) and slow disease progression. Several other phenotypes, including hyperCKemia, scapuloperoneal syndrome and distal myopathy with anterior tibial onset have also been reported in association with dysferlin mutations. HyperCKemia is an unusual clinical entity in which patients have elevation of creatine kinase in the absence of overt weakness or other muscle-related symptoms.

Table 1: Dysferlinopathies [adapted from Aoki (68)]

Subtype	Age of onset (years)	Clinical features	Prevalence
LGMD2B	26 ± 9	Slowly progressive proximal muscle weakness	20% of all AR LGMD
MM	21 ± 7	Slowly progressive distal weakness, legs > arms, calves most affected	UNK
Scapuloperoneal syndrome	?	Shoulder girdle, distal legs	Rare
Distal myopathy/anterior tibial onset	?	Distal weakness, anterior instead of calves	Rare
HyperCKemia	?	None (occasional calf hypertrophy)	UNK

UNK, unknown.

Dysferlin function

Dysferlin is a member of the ferlin family, a group of related proteins homologous to the *Caenorhabditis elegans* gene Fer-1 (11). All ferlins are characterized by multiple calcium-binding C2 domains (12). In mammals, dysferlin and myoferlin are the major ferlins expressed in skeletal muscle (13). Dysferlin is highly enriched at sites of muscle membrane injury, and the primary role for dysferlin is in calcium-mediated membrane repair (12). Dysferlin appears to facilitate the fusion and incorporation of membrane vesicles at the site of membrane discontinuity (14). The mechanism of action through which it promotes this process is still being elucidated, although it likely involves the ability of dysferlin to initiate multiple protein–protein and protein–phospholipid interactions (15). Dysferlin is also important for myoblast fusion during muscle development. However, myoferlin appears to be the ferlin most involved in this process (16).

Dysferlin pathogenesis

Muscle membranes are subjected to dramatic changes during contraction and are believed to incur microdomain injuries. These injuries are continually repaired by incorporation of new membrane vesicles. Mutations in dysferlin lead to decreased levels of dysferlin protein (6), and lack of dysferlin likely leads to an inability to repair these membrane injuries (14). This in turn leads to the muscle breakdown associated with the dysferlinopathies. This presumed pathogenic sequence is supported by the observation in patient biopsies of injured muscle membranes with accumulated unfused submembranous vesicles (17). It is also corroborated by the observation that dysferlin mutant mice develop a severe, progressive muscle disease (14). Interestingly, the levels of dysferlin protein do not appear to correlate with disease severity (18). Additionally, individual mutations can be associated with both LGMD2B and MM (19). Thus, other factors including modifier genes and physiological features specific to different muscle groups are predicted to determine the clinical presentation of dysferlin deficiency.

Caveolin-3

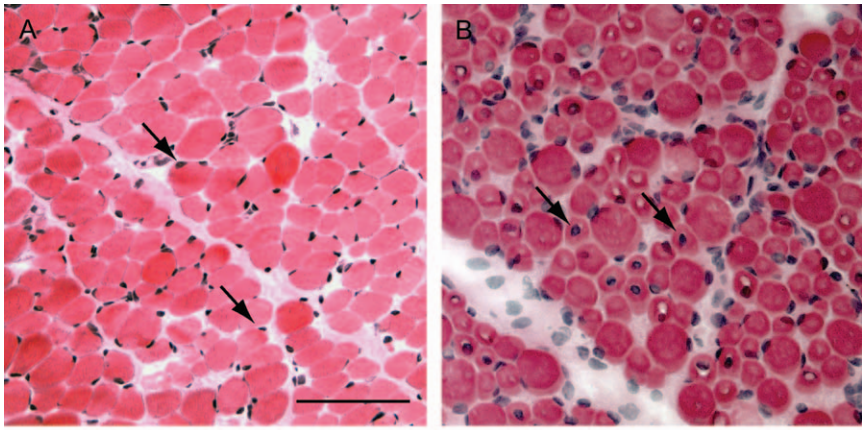
Caveolinopathies

Mutations in caveolin-3 (CAV-3) cause four distinct but overlapping forms of muscle disease (7). The first characterized was autosomal dominant LGMD 1C, which is a classic limb girdle dystrophy in that it features progressive weakness and dysfunction of the proximal musculature (20). LGMD1C most commonly presents in childhood, and is associated additionally with myalgias (muscle pains), muscle cramps, and high CK levels. Mutations in cav3 also cause autosomal dominant rippling muscle disease (RMD). The clinical features of RMD (percussion-induced rapid contraction, painful percussion-induced muscle mounding, and stretch/mechanically-induced muscle rippling) can be present in any combination in patients with CAV-3 mutations. The age of onset is widely variable (early childhood to 5th decade). Usually there is a history of exercise-induced stiffness and cramps and calf hypertrophy, but there is some irregularity in the presence of these features. Biopsy often shows variable fiber size and centralized nuclei. The remaining two

Table 2: Caveolinopathies [adapted from Bruno et al. (69)]

Subtype	Age of onset	Clinical features	Prevalence
LGMD1C	Childhood	Slowly progressive proximal muscle weakness, myalgias/cramps	Rare (<1% of LGMDs)
RMD	Usually late childhood	Percussion-induced muscle mounding, percussion-induced rapid contraction, mechanically induced muscle rippling	Rare
Distal myopathy	Early adulthood	Hand/foot muscle atrophy, elevated CK	Single case
HyperCKemia	?	None	UNK

UNK, unknown.



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Genetic causes of centronuclear myopathy

1. X-linked
   - Myotubularin (MTM1)
2. Autosomal Dominant
   - Dynamin-2 (DNM2)
3. Autosomal recessive
   - Amphiphysin-2 (BIN1)
4. Sporadic
   - hJumpy
   - Ryanodine receptor (RYR1)

Figure 1: Centronuclear myopathy. A, B) Shown are photomicrographs from hematoxylin/eosin-stained muscle from an unaffected child (A) and a child with myotubular myopathy (B). Note the small fiber size and the abundant central nuclei (arrows). (Images courtesy of Jeff Golden, University of Pennsylvania). C) List of the genes associated with centronuclear myopathy. Scale bar = 50 μm.

conditions associated with CAV3 mutations are hyperCKemia and distal myopathy. CAV3 mutations have been found in sporadic and dominant familial cases of hyperCKemia. Distal myopathy as a result of CAV3 mutations is rare and, as the name implies, characterized by distal weakness, as opposed to the proximal weakness observed in LGMD.

CAV-3 function

Caveolins are the core proteins associated with caveolae (21). Caveolae are 60–80 nm plasma membrane invaginations that mediate the sequestration of certain receptors and transporters and delivery of their cargo to the endocytic pathway. Caveolins and caveolae are also implicated in the regulation of lipid homeostasis, cell adhesion and cell signaling. There are three caveolins, with CAV-3 as the major muscle-specific isoform (22). Caveolae are intimately associated with the T-tubule network during myogenesis (23). In mature muscle, CAV-3 is found in the sarcolemma, not the T-tubule, and is also found at the neuromuscular junction (24). The function of CAV-3 has been defined using gene knockout in mice and gene knockdown in zebrafish (25,26). CAV-3 knockout mice lack caveolae in their muscle and have aberrant formation and organization of their T-tubule network. The knockout mice also experience myofiber degeneration, although the basis for this is poorly understood. Zebrafish with reduced levels of CAV-3 have abnormalities in myoblast fusion, a function for caveolin further supported by *in vitro* studies, as well as myofiber breakdown.

CAV-3 pathogenesis

The CAV-3 mutations associated with human muscle diseases likely act in a dominant-negative manner. A number of studies have demonstrated that there is a greater than 95% reduction in CAV-3 at the plasma membrane in patients with CAV-3 mutations (7,27). The mechanism behind this appears to be retention of both mutant and wild-type CAV-3 in the Golgi apparatus, with the result being the failure of trafficking of cav-3 to the plasma membrane. Mouse knockout and zebrafish knockdown studies (see above) suggest that reduced cav-3 levels can cause muscle disease. However, how the loss of caveolae results in the specific phenotypes observed in LGMD and RMD is still unclear. T-tubule abnormalities and calcium dysregulation may help explain the hyperexcitability and muscle rippling observed in these conditions. Interestingly, CAV-3 mutations also result in dysferlin mislocalization to the Golgi apparatus (28). Thus, it is possible that some or most of the pathophysiology in the caveolinopathies, in particular in LGMD1C, may be related to loss of proper dysferlin function.

Centronuclear Myopathies

Centronuclear myopathies are a diverse group of inherited muscle diseases that share a common pathologic appearance on muscle biopsy. Muscle fibers are characterized by enlarged, centrally located myonuclei surrounded by a halo of disorganized cytoplasmic organelles (Figure 1B). This is in contrast to normal myofibers, where nuclei are at the periphery and organelles are in a narrowly restricted location (Figure 1A). It is also distinct in appearance from the centralized nuclei observed in the muscular dystrophies. The abnormal fibers in centronuclear myopathy resemble fetal myotubes, although recent evidence suggests they accumulate after myogenesis is concluded (see below).

Centronuclear myopathies can present sporadically or else be inherited in X-linked, autosomal dominant and autosomal recessive patterns (Figure 1C) (8). The age of onset and the severity of presentation differ between the different types. The X-linked and autosomal recessive forms typically present in infancy with severe symptoms, while most cases of autosomal dominant centronuclear myopathy present in adulthood. Currently, four genes are known to cause the condition, with several cases not yet explained (and thus implying the existence of additional disease genes). Interestingly, despite the differences in clinical presentation, all known causes of centronuclear myopathy result in the same pathologic changes and are because of abnormalities in proteins associated with membrane traffic. The proposed function(s) of these genes as well as the possible relationship between these proteins is discussed below.

Myotubularin

Myotubular myopathy

Myotubular myopathy is an X-linked congenital myopathy caused by mutations in the myotubularin gene (29). It is one of the most severe muscle diseases of childhood, with symptom onset at or around the time of birth. Clinical characteristics include facial and eye muscle weakness, diffusely low muscle tone and respiratory failure as a result of diaphragmatic muscle dysfunction. Two-thirds of male patients die in the first year of life. The remaining boys often have significant disability. Classic myotubular myopathy is a rare condition, with estimated incidence approximately 1:50 000. Improved genetic testing, however, has demonstrated that muscle disease associated with myotubularin mutations is likely to involve significantly more clinical presentations (including late onset muscle weakness in women) with a much higher overall incidence (30–32).

Myotubularin function

Myotubularin was first described as a protein tyrosine phosphatase, but it is now clearly established that the primary function of myotubularin is as a lipid phosphatase that acts on phosphoinositide (PI) residues (32–34). Specifically, it dephosphorylates phosphoinositide-3-phosphate (PI3P) to form phosphoinositide monophosphate and dephosphorylates phosphoinositide-3,5-phosphate to form phosphoinositide-5-phosphate (PI5P). PIs are specialized lipid residues found restricted to various subcellular organelles (33). Their role is to target the localization of proteins to these organelles, and their modification/regulation is a critical aspect of regulated membrane traffic. The fact that myotubularin is a PI phosphatase thus implicates it as a regulator of membrane traffic.

Myotubularin has been studied extensively *in vitro*. Exogenous expression of the protein reduces PI3P levels, thus supporting the notion that it is a lipid phosphatase (34,35). Overexpression results in abnormalities in receptor endocytosis and endosome to lysosome trafficking and in the pathologic accumulation of endosomal vesicles (36). This phenotype is consistent with myotubularin's ability to dephosphorylate PI3P, which is itself a critical regulator of endosomal dynamics, and leads to the theory that myotubularin's major function is to regulate the movement of vesicles from the endosome to the lysosome (37). Myotubularin localizes in cell culture to early and late endosomes. It also localizes (perhaps more abundantly) to a dense cytoplasmic network of unclear significance and can dynamically associate with specialized plasma membrane structures like Rac-induced membrane ruffles (38). It is thus possible that myotubularin has additional non-endosomal functions, such as the prevention of inappropriate accumulation of PI3P on other organelles/structures or the regulation of plasma membrane dynamics by quickly removing PI3P. Other potential functions for myotubularin relate to PI5P (39). Phosphatase activity by myotubularin and its family members is the major mechanism for generating this PI. This PI is of low abundance and its function is poorly understood. A recent study suggests that PI5P has a role in the regulation of the PI3 kinase/Akt pathway (40).

Myotubularin pathogenesis

An essential role for myotubularin has been confirmed by a mouse knockout experiment, giving insight into how myotubularin deficiency leads to human muscle disease. Buj-Bello et al. found that loss of myotubularin causes a myotubular myopathy-like disease in mice (41). Their study provides convincing evidence that the defect is related to the maintenance of muscle and not its formation. Myogenesis is successfully completed in the myotubularin knockout mouse, and the pathognomonic changes associated with myotubular myopathy do not surface until the mice are at least 2 months old.

While this study shows that myotubularin is essential for post-development muscle maintenance, to date there have been no studies *in vivo* testing the mechanistic

hypotheses that have been generated by the *in vitro* work. Loss of regulation of endosome to lysosome traffic remains the predominant hypothesis. However, no vacuoles or endosomal accumulations have been detected in muscle biopsy samples, nor in myocytes cultures from myotubular myopathy patients. Furthermore, vacuole formation is not a described feature of other human diseases caused by mutations in myotubularin-related proteins. In contrast, it was recently reported that abnormalities in Fig4 and Vac14, two regulators of lipid monophosphates that act on the 5 phosphate residue, cause neurologic disease and result in a massive accumulation of pathologic vesicles (42,43). Thus, vesicle accumulation can result from dysregulation of lipid monophosphates, and why this feature is absent in cells with myotubularin deficiency remains to be explained if the endosomal trafficking hypothesis is correct.

Dynamin-2

Autosomal dominant centronuclear myopathy

Mutations in the large GTPase dynamin-2 are associated with autosomal dominant centronuclear myopathy (44). In general, cases of dynamin-2-related centronuclear myopathy are late in onset, with symptoms first appearing in adolescence or adulthood. The disease is characterized by a slowly progressive weakness, with distal muscles more prominently affected than proximal (45). Facial weakness is a common feature, and some patients exhibit bilateral ptosis and extraocular eye muscle involvement. No reduction in life span is associated with the condition. Recently, additional mutations in dynamin-2 have been identified in patients with a more severe form of centronuclear myopathy resembling myotubular myopathy (46). These patients present with muscle weakness at birth. In contrast to myotubular myopathy, however, these infants have little or no respiratory involvement, and have a relatively good prognosis.

Dynamin-2 function

Dynamin 2 is a conventional member of the dynamin superfamily, sharing particular homology with dynamin 1 and 3. Dynamins 1-3 share a similar structure, with 5 conserved functional domains [GTPase, pleckstrin homology (PH), middle, proline rich and GTPase effector (GED)] (47). Dynamins 1 and 3 are restricted in their tissue distribution, while dynamin-2 is ubiquitously expressed (48). Dynamins are implicated in the regulation of a variety of cell and organelle fission events (49). Dynamin-2, in particular, has long been known to play a key role in clathrin-mediated endocytosis, thought to function by forming a ring around vesicle necks and contribute to the 'pinching' off of vesicles. Recent observations suggest that dynamin 2 also functions in a number of cellular processes not directly related to its role in vesicle fission. In particular, dynamin 2 interacts with several proteins involved in the regulation of actin assembly, including cortactin, syndapins, intersectins, and profilin (50). These interactions likely assist in coordinating endocytosis, but they also may contribute to cell functions such as lamellipodial extension (51), phagocytosis (52), and cell motility (53). Dynamin-2 is also a critical player in cytokinesis and regulation of centrosomal function.

Dynamin-2 pathogenesis

To date, all muscle disease caused by mutations in dynamin-2 are confined to the middle and PH domains (Figure 2A). These mutations are believed to act in a dominant-negative manner as limited study of dynamin-2 in patient fibroblasts revealed no decrease in protein levels (44). Current understanding of these domains suggests that the phenotypes caused by mutations in either domain may result from defective endocytosis. There is evidence that the middle domain is necessary for assembling dynamin into the ring-like structures surrounding the necks of endocytic vesicles (54). The middle domain also interacts directly with the GED (55), which may be the basis for this

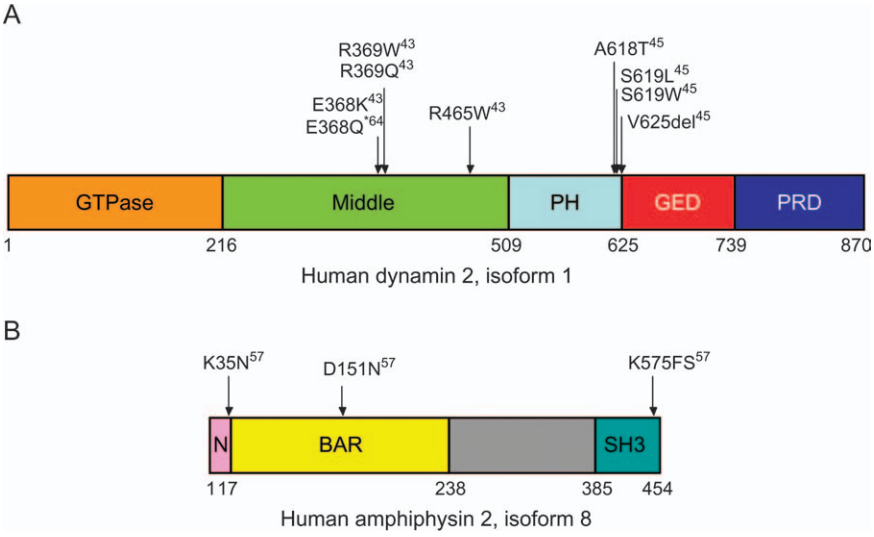

Figure 2: Mutations in centronuclear myopathy. A) List of mutations in dynamin 2. Note that all reported sequence changes are confined to the middle and PH domains. Note that the E368Q mutation* causes both myopathy and peripheral neuropathy (67). B) List of mutations in amphiphysin 2 (see text for more description).

requirement in dynamin self-assembly. While the PH domain of dynamin is not known to have a direct role in self-assembly, it belongs to a family of domains found in a wide variety of membrane-associated proteins. Many PH domains bind PIs with high affinity and specificity. Dynamin's PH domain binds to membrane phospholipids including PI(4,5)p2 and PI(3–5)P3 (56). Mutations disrupting phosphoinositol binding of the PH domain of dynamin-1 block endocytosis (57), which suggests that endocytosis might also be disrupted in dynamin-2 PH domain mutations.

Little is known about the specific functions of dynamin-2 in muscle, and the reasons dynamin-2 mutations cause myopathy remain unknown. Middle domain mutations have only been reported in the more common, less severe presentation, while the PH domain mutations are associated with a severe presentation; it is not clear why this distinction exists. The original study identifying dynamin-2 mutations in centronuclear myopathy also examined the consequence of one point mutation on dynamin-2 localization (44). They found that mutant dynamin-2 is aberrantly expressed, and postulate that the mutant protein acts in a dominant-negative fashion by preventing the proper transport and localization of the wild-type protein. This is an intriguing hypothesis; however, no changes in dynamin-2 levels or localization have been found in fibroblasts or skeletal muscle from patients.

Amphiphysin 2

Autosomal recessive centronuclear myopathy

Recently, the first gene associated with autosomal recessive centronuclear myopathy was identified (58). Using a candidate gene approach, Laporte and colleagues found mutations in amphiphysin 2 (BIN1) in four individuals from three families with parental consanguinity and presumed recessive inheritance. The four patients had onset of weakness in infancy, and the classic features of centronuclear myopathy on muscle biopsy. Other features, including facial and eye muscle weakness, were variable. Respiratory failure was reported only in a fifth individual with a suspected BIN1 mutation. In all, the phenotype more closely resembles that reported for the severe dynamin-2 mutations than that of myotubular myopathy.

Amphiphysin 2/BIN1 function

Amphiphysin-2 is part of larger family of proteins containing a common structural domain called the BAR (Bin1/amphiphysin/RVS167) domain. The BAR domain dimerizes to form an arched structure that can readily bind to curved membranes (59). BAR domain proteins are membrane adaptors whose fundamental function appears to be as sensors of membrane curvature (60). This function is utilized during late endocytosis, when amphiphysins bind to the budding clathrin-coated vesicle and recruit various proteins, including dynamin (61). The interaction with dynamin occurs via amphiphysin's SH3 domain. In addition to regulating endocytosis, amphiphysins are involved in a diverse range of cellular processes including signal transduction, transcriptional regulation, apoptosis and vesicle fusion (60).

Amphiphysin-2 is a ubiquitously expressed but has several different tissue-specific isoforms. The muscle-specific isoform, called M-amphiphysin-2, is highly expressed during myogenesis and in adult muscle and localizes to membrane subdomains including T-tubules (62). It is unique among amphiphysin-2 isoforms in that it lacks a domain that is important for binding clathrin-coated vesicles and regulating endocytosis, and instead contains a novel exon (exon 10) that promotes association with PI(4,5)P2 at the plasma membrane. De Camilli and co-workers, in a seminal study published in 2002, discovered that M-amphiphysin is able to independently promote tubule evagination, a function utilized *in vivo* for the biogenesis of T-tubules (63). Knockdown of M-amphiphysin-2 in C2C12 myoblasts prevents myoblast fusion (63), while gene knockout in mice results in a severe, lethal cardiomyopathy (64).

Amphiphysin pathogenesis

The mechanisms underlying amphiphysin dysfunction are the best understood among the centronuclear myopathies. An extensive study by Laporte and colleagues, building on their previous work on dynamin-2 as well as the studies by De Camilli and others, identified the disease-causing mutations in the amphiphysin-2 gene and discovered important clues about their pathogenesis (Figure 2B) (58). Two of the reported mutations occur in the BAR domain and cause the elimination of the ability of the protein to promote tubulation *ex vivo*. The other two mutations occur in the SH3 domain. The resulting proteins are unable to interact with dynamin-2 and in turn prevent the association of dynamin-2 with the T-tubule network. The authors thus hypothesize that mutations in amphiphysin-2, either by directly interfering with its tubulation function or by eliminating the important interaction with dynamin-2, disrupt the formation and maintenance of the T-tubule network. This study found via immunostaining of muscle biopsies that the network is, in fact, disrupted. What is not clear, however, is whether alteration in the tubule network is what causes the muscle pathology in patients with centronuclear myopathy. In addition, as the authors point out, this mechanism does not explain how amphiphysin dysfunction results in the most characteristic pathologic change, the development of central nuclei. This is also true for dynamin-2 and myotubularin, where involvement in the trafficking pathways cannot explain the formation of the very unusual appearance of the muscle. Whether the centronuclear phenotype results from a novel functional pathway of these proteins, or whether instead it is an epiphenomenon of disease, remains to be determined.

Sporadic Centronuclear Myopathies

Many cases of centronuclear myopathy occur without an obvious pattern of inheritance. Within those cases, some

boys have mutations in myotubularin. The remaining cases have been difficult to classify genetically. Mutations in two additional genes have been reported in individual patients with centronuclear myopathy. One is in the ryanodine receptor (RYR1), a gene important for the regulated release of calcium stores (65). Abnormalities in RYR1 are more commonly associated with another congenital myopathy called central core disease. The other is the hJumpy gene. Two patients have been found with mutations in this gene, which was uncharacterized prior to the mutation report (66). The authors who discovered the mutations determined that hJumpy is a lipid phosphatase with enzymatic activity identical to that of myotubularin, and that the observed sequence changes reduce hJumpy's phosphatase activity *in vitro*. The gene otherwise does not share myotubularin's other functional domains and is not a member of the myotubularin family. This finding again demonstrates that myotubularin-like phosphatase activity is critical for muscle function and for disease pathogenesis in centronuclear myopathy. Further work needs to be carried out to establish hJumpy's role *in vitro* and *in vivo*. In addition, the relationship between hJumpy and myotubularin is unknown; it is not clear if they have overlapping functions and act on the same pool of PIs, or if instead they have different spatial and temporal functions.

Summary

At the present time, mutations in six components of the membrane traffic machinery have been identified that result in muscle disease. They can be broken down broadly into two groups, those causing LGMD and those causing centronuclear myopathy. Given that mutations in three of the six were only discovered in the past 2 years, we are likely only scratching the surface in terms of the association between membrane traffic and muscle function and pathology. Currently, no common theme or definite pathway that links all six genes has been uncovered. The finding of dysfunction of the T-tubule network in several of the cases suggests there may be a common pathway in which these components are involved. Furthermore, it underscores the importance of membrane traffic for T-tubule biogenesis and homeostasis, although it is still unclear how T-tubule biology is an important aspect of the pathogenesis or why membrane traffic is so important for muscle function. It seems likely, as parallel studies on the fundamental cell biology of trafficking and muscle merge with human genetics, that a deeper understanding of the normal and pathologic processes will emerge.

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