The protein synthesis and degradation of eukaryotic cells must be highly selective and tightly regulated to maintain cellular homeostasis. Like other tissues, muscle contains multiple pathways for protein breakdown including the lysosomal, Ca^{2+}-dependent, and cytosolic ATP-dependent and independent proteolytic systems. These pathways are considered to play important roles not only in cellular degeneration and death but also in cellular differentiation, growth and regeneration. In fact, many lysosomes appear in the regenerating skeletal muscle fibers (1), and they disappear as the myofibers mature.

Distal myopathy with rimmed vacuole formation (DMRV) is a sporadic or an autosomal recessively inherited disorder characterized by the formation of rimmed vacuoles in skeletal muscle fibers. A rimmed vacuole is believed to be an autophagic vacuole in which activation of lysosomal proteases such as cathepsins B, L, or H have been reported (2, 3). Although the lysosomal system is not well developed in the differentiated mature myofiber, these cathepsins in DMRV are thought to derive from myofiber itself, not from macrophages.

On the other hand, a calcium-dependent neutral proteinase has been suggested to be involved in the muscle degradation in Duchenne muscular dystrophy (DMD) (4, 5), in which dystrophin, a sarcolemmal protein, is deficient (6). Calpain is the non-lysosomal enzyme which is activated by a high concentration of Ca^{2+} ion with a neutral pH condition. Instability of sarcolemma caused by dystrophin deficiency in DMD might result in the influx of a high concentration of extracellular calcium ion which activates intracytoplasmic calpain. Socalpain might play an important role in the initiation of myofibrillar degeneration in muscular dystrophies including DMD. However, this plausible hypothesis explaining the myofibrillar destructive process in DMD includes some discrepancies. For example, the optimal Ca^{2+} concentration for calpain activation is much higher than the actual Ca^{2+} concentration in muscle cells from DMD patients, and furthermore, the opaque fiber which is presumed to contain a high concentration of Ca^{2+} ion was immunohistochemically negative for calpain (5). Further studies will clarify these points. Recently, mutation of calpain 3 gene was reported to be causative in limb-girdle muscular dystrophy type 2A (LGMD 2A) (7). This is the first case in which gene encoding functional protein, not cytoskeletal protein, is responsible for muscular dystrophy. In DMRV, Kumamoto et al (8) reported the increased stainability of calpain around the rimmed vacuoles or in the cytoplasm of myofibers. Therefore both lysosomal and Ca^{2+}-dependent proteolytic pathways contribute to the pathogenesis of DMRV.

Skeletal muscle also contains the soluble ATP-ubiquitin-dependent proteolytic system. Ubiquitin is a small 76-amino acid molecule that is highly conserved across mammalian evolution (9). Ubiquitin is one of the cell’s heat-shock proteins (HSP) which are induced by heat or by other types of stress. The expression of ubiquitin might be the safeguard mechanism to eliminate abnormal proteins which are synthesized abundantly at the time of stress. Degradation of protein via this pathway involves two distinct steps: first, signaling of the protein by covalent attachment of multiple ubiquitin molecules using three enzymes, El (ubiquitin-activating enzyme), E2 (ubiquitin-conjugating enzyme), and E3 (ubiquitin ligase). The second step is the degradation of the targeted protein by an ATP-dependent protease, 26S proteasome. The first process of linking ubiquitin to lysine residues of proteins destined for degradation involves the activation of ubiquitin by the E1 enzyme in an ATP-dependent manner. Activated ubiquitin is then transferred to an E2 carrier protein and then to the substrate protein, a reaction catalyzed by an E3 enzyme. The process is repeated as multiple ubiquitin molecules are added to form a ubiquitin chain. This modification of the substrate leads to its rapid degradation of targeted protein by a very large proteolytic complex, the 26S proteasome which requires ATP to function. The striking features of this pathway are its rapidity, specificity, and efficiency in the degradative process of proteins. Other than the function of presenting the degenerating signals, recent studies have defined that ubiquitin has multiple functions, such as ubiquitination of activated T-cell antigen receptor (10) or ubiquitination of the peptide processed for antigen presentation on MHC class I molecules (11). This system is widely accepted to catalyze the rapid breakdown of highly abnormal polypeptides and also to participate in the breakdown of various short-lived normal proteins like cyclin, the cell cycle associated protein (12). In addition, Medina et al (13) reported that this pathway degrades muscle proteins including the long-lived myofibrillar components such as actin and myosin in physiologic and some pathologic conditions. They also reported activation of the ubiquitin-proteasome proteolytic pathway in muscle atrophy caused by starvation and denervation (13).

Recent studies have clarified the accumulation of ubiquitin in and around the rimmed vacuoles in both DMRV and inclusion body myositis (14). Interestingly, the vacuoles in DMRV also contain Congophilic amyloid materials and deposits which are immunoreactive for β-amyloid protein, β-amyloid protein precursor, and hyperphosphorylated tau protein, in addition to ubiquitin, as reported in the brain of Alzheimer’s patients (15).
Thus accumulation of ubiquitin may not be disease specific and the common pathogenic mechanism for the accumulation of ubiquitin and the other proteins is considered among these diseases. Therefore, the significance of this system in DMRV has not been defined.

Kumamoto et al in this issue (16) examined the role of ATP ubiquitin-dependent proteolytic pathway in the myofiber degradation of DMRV.

See also p 746.

They found localization of proteasome around the vacuoles and in the atrophic muscle fibers. Ubiquitin was localized in the rimmed vacuoles, and less in the cytoplasm. According to their findings, the ubiquitin-proteasome pathway plays some role in the protein degradation in DMRV, although the lysosomal pathway may be the main pathway for protein degradation in this myopathy.

At present, the relationship between the ubiquitin-proteasome pathway and the other proteolytic systems, i.e. lysosomal and Ca2+-dependent pathways, is uncertain. With regard to this important issue, Doherty et al (17) report the evidence that ubiquitin is accumulated in lysosomes. Kumamoto et al (16) also mentioned in their article that ubiquitinated protein might be readily taken up into lysosomes. On the other hand, they (8) suggested that partially disrupted myofibrils in the extralysosomal system, i.e. calpain, in an earlier stage might be degraded completely in rimmed vacuoles in DMRV. However, differing from DMD in which the influx of Ca2+ from extracellular fluid activates the Ca2+-dependent pathway, the trigger for activation of calpain in DMRV is uncertain. Further works dealing with these issues in depth and the initial trigger for the protein degradation in DMRV will shed light on the pathogenesis of DMRV and contribute to the development of new medical treatments for DMRV.

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