these mice, BIN1 and T-tubule defects preceded the appearance of muscle weakness. Similar BIN1 and T-tubule/triad defects appear as common features of the different CNM forms due to mutations in *MTM1*, *BIN1*, or *DNM2* in humans, suggesting that these proteins functionally interact. In agreement, Dowling et al. [11] recently showed that DHPR and RYR1 labeling was aberrant in three XLMTM patients. In both *mtm1* zebrafish morphants and in our present study on CNM patients, electron microscopy showed some defects in the triad structure. BIN1 accumulations around or between centralized nuclei were observed in patients with *BIN1* and *MTM1* mutations (Figs. 5, 6). Surprisingly, triad and BIN1 defects were marked in *DNM2*-mutated patients' biopsies, where DHPR, RYR1, and BIN1 were all distributed in a longitudinal orientation in contrast to their typical transverse pattern in normal skeletal muscle. As *DNM2*-mutated patients studied here are adults, these defects may have accumulated with time, suggesting the implication of DNM2 in the maintenance of triad organization.

Taken together, our findings suggest that the three forms of CNM share a common pathogenetic mechanism where BIN1 may represent a molecular link between myotubularin and dynamin 2 in skeletal muscle. *MTM1* encodes a PI phosphatase, and the skeletal muscle-specific exon of *BIN1* encodes a PI-binding domain. Moreover, BIN1 and DNM2 are well-known interactors in cultured cells although this molecular link remains to be explored in skeletal muscle. We hypothesize that MTM1 regulates the level and localization of specific PIs that either bind to BIN1 or serve as substrates to produce the PI that specifically binds BIN1. Amphiphysins have been shown to bind preferentially to PtdIns(4,5)P_2 rather than substrates and product of myotubularin activity (PtdIns3*P*, PtdIns(3,5)P_2, and PtdIns5*P*) [5, 21, 24, 43, 46]. Once membranes are remodeled by the action of BIN1, DNM2 may direct the adequate organization of the triads and/or their maintenance through cytoskeleton regulation. Indeed, DNM2 is a microtubule-binding protein which also plays a role in actin cytoskeleton assembly [42, 48]. An alternative hypothesis is the participation of BIN1 for the delivery of DHPR ion channels to the T-tubules as suggested recently by Hong et al. [17] based on their results in cardiac myocytes. Whether this mechanism is present in skeletal muscle remains to be determined.

In conclusion, BIN1 skeletal muscle isoforms appear to play an important role in triad formation, and this function is altered in several forms of centronuclear myopathies, where BIN1 and triad organization defects define a common pathogenetic mechanism. Molecular dissection of the roles of BIN1, myotubularin, and dynamin 2 in skeletal muscle will be required to understand the precise regulation of this pathway. Specifically, it will be important to elucidate what other functions BIN1 has in addition to its known role as a tumor suppressor.

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Myonuclear breakdown in sporadic inclusion body myositis is accompanied by DNA double strand breaks

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Abstract

Rimmed vacuoles in sporadic inclusion body myositis (s-IBM) contain nuclear remnants. We sought to determine if the nuclear degeneration seen in s-IBM is associated with DNA damage. In muscle biopsy specimens from ten patients with s-IBM and 50 controls, we immunolocalized 1) phosphorylated histone H2AX (γ-H2AX), which is a sensitive immunocytochemical marker of DNA double-strand breaks and 2) DNA-PK, which is an enzyme involved in double-strand break repair. In s-IBM, vacuolar peripheries often showed strong immunoreactivity to γ-H2AX and the three components of DNA-PK (DNA-PKcs, Ku70, and Ku80). A triple fluorescence study of Ku70, emerin, and DNA displayed nuclear breakdown and it suggested impaired nuclear incorporation of Ku70. The percentage of positive nuclei for γ-H2AX was significantly higher in vacuolated fibers than non-vacuolated fibers in s-IBM, or fibers in polymyositis. We hypothesize that a dysfunction of nuclear envelope may cause nuclear fragility, double-strand breaks and impaired nuclear transport in s-IBM.

Keywords: Inclusion body myositis; Rimmed vacuole; DNA double strand breaks (DSB); Nuclear breakdown

1. Introduction

Sporadic inclusion body myositis (s-IBM) is the primary cause of acquired myopathy in patients over 50-years old, but no effective therapy has yet been found [1,2]. The histopathological hallmarks of s-IBM consist of mononuclear cell infiltration, muscle fibers with congophilic inclusions, and rimmed vacuoles. Several studies showed nuclear components in the rimmed vacuoles (e.g., a single-stranded DNA binding protein of nuclear origin [3], emerin [4,5], lamin A/C [4] and histone H1 with DNA [5]). The findings indicate that the vacuoles may result from nuclear breakdown.

Terminally differentiated cells do not possess a replication-associated DNA repair mechanism, making them particularly sensitive to DNA damage [6]. Mature muscle cells are such terminally differentiated cells. In a muscle cell culture study, the exposure of differentiated myocytes to hydrogen peroxide resulted in the accumulation of foci of phosphorylated histone H2AX (γ-H2AX) [7], which is a sensitive marker of a serious form of DNA damage, DNA double strand breaks (DSB) [8]. DSB are produced by reactive oxygen species (ROS), ionizing radiation, and other genotoxic agents. Histone H2AX, a variant of histone H2A, is rapidly phosphorylated at Ser 139 in the chromatin region surrounding a DSB [9]. Immunocytochemical staining of γ-H2AX has been broadly applied to reveal DNA damage caused by cancer and other cellular stresses [8,10]. DNA-PK is an enzyme involved in the initial step of

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the DSB repair process, non-homologous end joining (NHEJ), which does not require DNA replication, and therefore NHEJ is the predominant DNA repair mechanism in terminally differentiated cells [11,12]. DNA-PK consists of a catalytic subunit (DNA-PKcs) and two regulatory subunits (Ku70 and Ku80). The binding of heteroduplexes of Ku70 and Ku80 to DSB sites initiates the repair process [13,14]. In the current paper, we examine whether DSB are associated with myonuclear breakdown in s-IBM.

Note that DSB is different from the apoptotic DNA fragmentation that has been scarcely detected in the s-IBM muscles [15]. In DSB, DNA breaks occur directly and randomly by radiation or other genotoxic agents, whereas apoptotic DNA fragmentation takes place at a late stage of programmed cell death, in which endonucleases sever DNA strands at regular lengths. Apoptotic DNA fragmentation is not subject of repair or is not labeled with anti-γ-H2AX.

2. Materials and methods

2.1. Patients

We studied muscle biopsy specimens from 10 patients (58–82 years old, 8 men and 2 women) who fulfilled the clinical, electromyographic, and histopathological criteria for s-IBM [16]. The muscle sections displayed cell infiltration surrounding non-necrotic fibers, congophilic inclusions and rimmed vacuoles in each patient. All s-IBM patients showed slowly progressive muscular symptoms (disease duration: 3.8 ± 2.9 years; mean ± standard deviation [SD], range: 0.5–9 years). None of these patients had received immunotherapy before the muscle biopsy. Specimens from five patients without pathologic alterations served as non-pathologic controls. For controls of other neuromuscular diseases, we used 45 muscle biopsies from patients with polymyositis ($n = 10$), dermatomyositis (8), dystrophinopathy (3), dysferlinopathy (3), mitochondrial encephalomyopathy (5), myotonic dystrophy type I (1), neurogenic muscular atrophy (5), oculopharyngeal muscular dystrophy (5), myopathy with autophagic vacuoles of an undetermined etiology (1), rhabdomyolysis (1), hypokalemic vacuolar myopathy (2), and colchicine myopathy (1). The above diagnoses were based on a clinical examination, family history, electromyography, and muscle biopsy studies. Polymyositis and dermatomyositis were diagnosed using conventional criteria [17]. The polymyositis sections contained several to many non-necrotic fibers surrounded by mononuclear cells, and the dermatomyositis sections demonstrated perifascicular atrophy or perimysial infiltration of inflammatory cells. This study was performed with the compliance of the internal review board of our institution.

2.2. Immunohistochemistry

Table 1 shows the primary antibodies applied and their concentrations. Immunohistochemical studies were performed as previously described [18]. Briefly, sections were fixed in cold acetone and then in 4% paraformaldehyde in 0.1 M phosphate buffer (pH 7.4) before being blocked and incubated overnight at 4 °C with the primary antibody. The sections were then incubated with a biotin-labeled secondary antibody and developed using the avidin–biotin complex (ABC) immunoperoxidase method (Vector Laboratories, Burlingame, CA) with 3,3′-diaminobenzidine as the coloring agent. Next, the slides were lightly counterstained with hematoxylin for the quantitation of positive nuclei. The control experiments involved the omission of the primary antibody or the substitution of the primary antibody for non-immune mouse or rabbit IgG. We immunostained 12 or more sections from different individuals at a time, and the duration of color development was fixed. The specificity of antibodies for γ-H2AX and Ku70 was also tested in immunoblotting.

For triple-color immunofluorescence studies, the sections were incubated with anti-Ku70 plus anti-emerin antibodies followed by incubation with appropriate secondary antibodies for triple fluorescence (Chemicon International, Temecula, CA). The slides were mounted with Vectashield (Vector) containing 1.5 μg/mL of the nuclear DNA marker 4′,6-diamidino-2-phenylindole (DAPI) and examined with confocal imaging using the LSM510-META system (Carl Zeiss, Jena, Germany). As controls, we performed a single-color fluorescence study using each antibody or DAPI alone and confirmed the specificity of the secondary antibodies and filters.

Table 1
List of primary antibodies.

Antigen	Type	Clone/ID	Source	Concentration
γ-H2AX	MMA	JBW301	Upstate	1 μg/mL
Ku70	MMA	4C2–1A6	Abnova	1 μg/mL
Ku80	MMA	111	Abcam	1:500
DNA-PKcs	RPA	PC127	Calbiochem	5 μg/mL
Emerin	RPA	FL-254	Santa-Cruz Biotec	1 μg/mL
HNE	MMA	HNEJ-2	JaICA	20 μg/mL
iNOS	RPA	sc-651	Santa-Cruz Biotec	4 μg/mL
LAMP-2	MMA	H4B4	Santa-Cruz Biotec	4 μg/mL

MMA: mouse monoclonal antibody; RPA: rabbit polyclonal antibody.

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2.3. Immunoelectron microscopy

Preembedding immunoelectron microscopy was performed using the immunogold method with silver-enhancement [19]. Cryostat sections, prepared from s-IBM biopsy specimens that had been stored at −80 °C, were attached to a slide glass, fixed in 4% paraformaldehyde, and incubated with the anti-Ku70 antibody at a 100-fold dilution. The sections were then incubated with a secondary antibody (goat IgG, Fab' fragment) coupled with 1.4 nm gold particles (Nanoprobes Inc., Yaphank, NY). The sample-bound gold particles were then silver-enhanced using the HQ-silver kit (Nanoprobes) at room temperature for 12–14 min according to the manufacturer's instructions. Then, the samples were postfixed with 0.5% osmium oxide in 0.1 M phosphate-buffer at pH 7.4, before being dehydrated in a graded series of ethanol (50%, 70%, 90%, and 100%) and propylene oxide, and embedded in epoxy resin. Ultrathin sections were then cut, stained with uranyl acetate and lead citrate, and examined with a JEM-1400A electron microscope (JEOL Ltd., Tokyo, Japan).

2.4. Quantitation

For quantitation of the γ-H2AX-positive myonuclei in the non-pathologic controls, a mean of 342 randomly selected photographed nuclei were inspected for each sample. In s-IBM, a mean of 42.3 vacuolated fibers were photographed per patient. For each vacuolated fiber, we surveyed γ-H2AX-positivity in vacuoles and nuclei. To quantitate the positive nuclei in non-vacuolated fibers, we analyzed a mean of 266 nuclei in 97 randomly selected muscle fibers in each s-IBM patient. In polymyositis, we analyzed a total of 1657 randomly selected myonuclei. In

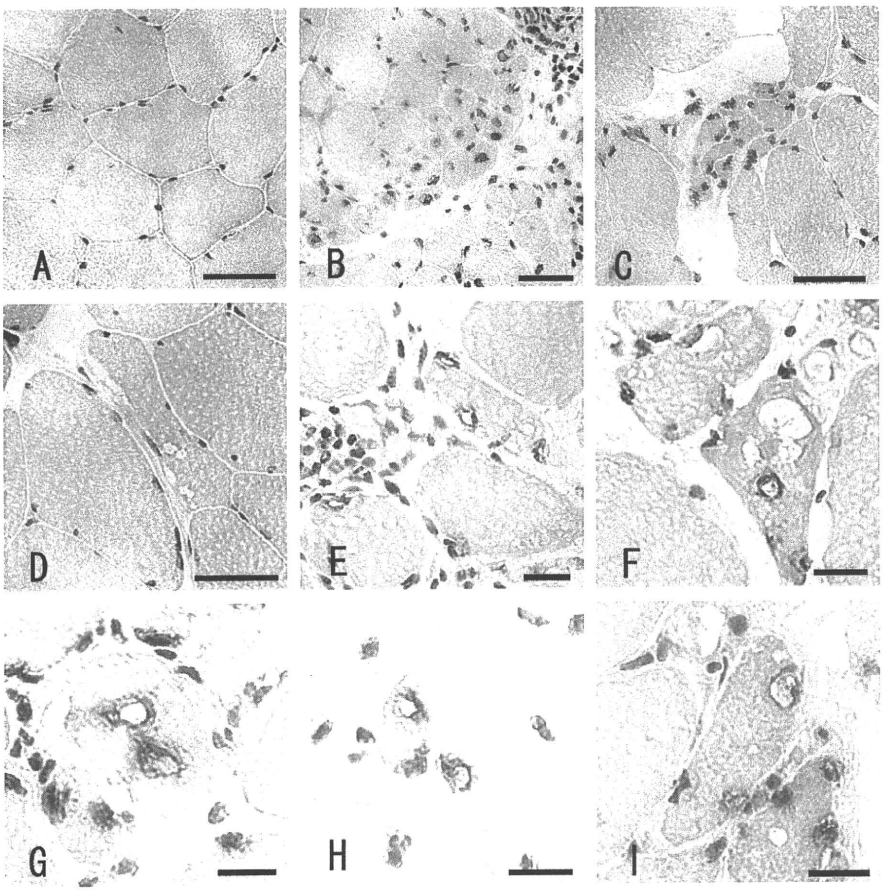

Fig. 1. A–F: Localization of γ-H2AX, which is induced upon the occurrence of DNA double-strand breaks (DSB); in controls (A–D); and sporadic inclusion body myositis (s-IBM) (E–F). Immuno-peroxidase method, lightly counterstained with hematoxylin to localize nuclei. (A) non-pathologic control, (B) perifascicular atrophy in dermatomyositis, (C) grouped atrophy in neurogenic muscular atrophy, and (D) oculopharyngeal muscular dystrophy (OPMD). The myonuclei in A show no or trace immunoreaction to γ-H2AX. The nuclei of the atrophic fibers in B and C are strongly positive for γ-H2AX. In D, the vacuoles in OPMD are negative for γ-H2AX. In E & F, the vacuolar rims and myonuclei in s-IBM contain strongly positive γ-H2AX products. Some mononuclear cells in the inflammatory exudates in E are positive. G–I: Localization of the three components of DNA-PK (G: Ku70, H: Ku80 and I: DNA-PKcs) in vacuolated fibers in s-IBM. Vacuolar peripheries and nuclei display positive immunoreactivity for each component of DNA-PK. Bar = 50 μm (A–D); 20 μm (E–I).

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s-IBM DNA-PKcs, we examined a total of 393 vacuolated fibers with 950 nuclei and 1953 myonuclei in non-vacuolated fibers. As it was sometimes difficult to differentiate between the nuclei of invading/surrounding mononuclear cells and myonuclei, we excluded muscle fibers surrounding inflammatory cells from the nucleus calculation. We categorized nuclei as positive when a brown color was clearly discernible against lightly-stained hematoxylin.

3. Results

3.1. Increased expression of the DNA double strand break (DSB) marker γ-H2AX in s-IBM vacuolated fibers

In the non-pathologic controls, a small number of myonuclei showed a weakly positive reaction to γ-H2AX ($n = 5$; $6.0 \pm 1.8\%$, mean ± standard deviation [SD]. Range: 3.97–8.05) (Fig. 1A). In polymyositis and dermatomyositis, the nuclei in regenerating fibers were positive for γ-H2AX. The nuclei in perifascicular atrophic fibers in cases of dermatomyositis were strongly positive (Fig. 1B), and positive myonuclei were also found in other fibers in polymyositis and dermatomyositis. A proportion of the cells in inflammatory exudates were positive for γ-H2AX. In neurogenic muscular atrophy, strongly reactive nuclei were usually found in atrophic angulated fibers (Fig. 1C), and the nuclei at pyknotic nuclear clumps showed increased reactivity for γ-H2AX. In other neuromuscular diseases, the nuclei at nuclear clumps such as those observed in myotonic dystrophy showed increased reactivity for γ-H2AX, and the nuclei in ragged-red fibers in mitochondrial encephalomyopathy and those of regenerating fibers in various myopathies were strongly positive for γ-H2AX. Vacuoles in hypokalemic myopathy, myopathy with autophagic vacuoles, colchicine myopathy, and OPMD were negative for γ-H2AX (Fig. 1D).

In s-IBM, a proportion of fibers contained vacuoles that were partially or entirely lined by positive immunoreactivity (Fig. 1E and F). Table 2 shows the percentage of (1) vacuolated fibers vs. total fibers and (2) fibers containing γ-H2AX positive vacuoles vs. total vacuolated fibers in patients with s-IBM ($n = 10$; $74.0 \pm 13.0\%$, mean ± SD). The nuclei in vacuolated fibers displayed strong γ-H2AX-positive reactivity, and the percentage of positive nuclei was significantly higher in vacuolated fibers than in non-vacuolated fibers (Table 1) ($p < 0.01$; paired Student's t-test). In polymyositis, the percentage of γ-H2AX-positive nuclei ($n = 10$; $23.3 \pm 7.4\%$, mean ± SD) was similar to that in the non-vacuolated fibers in s-IBM, but lower than that in the vacuolated fibers ($p < 0.01$; Student's t-test).

The results of immunoblotting using this anti-γ-H2AX antibody showed several positive bands including ubiquitinated forms of γ-H2AX (Fig. 2) [20].

3.2. Detection of the DSB repair enzyme DNA-PK in s-IBM

In s-IBM, all of the DNA-PK components (DNA-PKcs, Ku70, and Ku80) were found in vacuolar peripheries as well as being strongly expressed in nuclei, consistent with the results for γ-H2AX (Fig. 1G, H and I). As for DNA-PKcs, $70.6 \pm 14.0\%$ (mean ± SD) of vacuolated fibers contained positive vacuoles for DNA-PKcs. The percentage of positive nuclei for DNA-PKcs was significantly higher in vacuolated fibers than in non-vacuolated fibers ($61.7 \pm 10.6\%$, mean ± SD, vs. $32.5 \pm 10.2\%$: $p < 0.01$; paired t-test). Ku70 was often found to form several round or comma-shaped cytoplasmic inclusions in vacuolated fibers and other fibers. We confirmed the relative localization of Ku70, the nuclear envelope, and DNA in a triple-fluorescence study in five patients with s-IBM, five patients with polymyositis, and patients with other diseases. In polymyositis and other controls, Ku70 was confined to within the emerin boundary, even when the Ku70-signal was very intense (Fig. 3A). In vacuolated fibers in s-IBM, although Ku70 was often localized to the nuclei, it was also found in vacuolar peripheries, around the nuclei, and in the cytoplasm (Fig. 3B and C). In a few instances, cytoplasmic Ku70-positive granules were associated with nuclear frag-

Table 2
Quantitation.

s-IBM Pt number	Vacuolated fibers/total fibers (%)	γ-H2AX positive fibers/vacuolated fibers (%)	Positive nuclei in	
			Vacuolated fibers (%)	Non-vacuolated fibers (%)
1	18.6	87.2	64.0	24.5
2	21.0	82.9	64.0	29.0
3	5.0	69.6	64.5	39.4
4	4.2	76.5	70.3	23.8
5	7.2	89.5	81.4	38.2
6	23.0	76.8	65.0	22.1
7	6.6	47.1	58.8	8.5
8	7.6	74.6	56.5	25.2
9	4.2	58.1	58.2	24.9
10	7.6	78.3	63.9	21.6
Mean ± SD	10.5 ± 7.3	74.0 ± 13.0	64.7 ± 7.1*	25.7 ± 8.8

* The percentages for vacuolated fibers are significantly higher than those for non-vacuolated fibers ($p < 0.01$).

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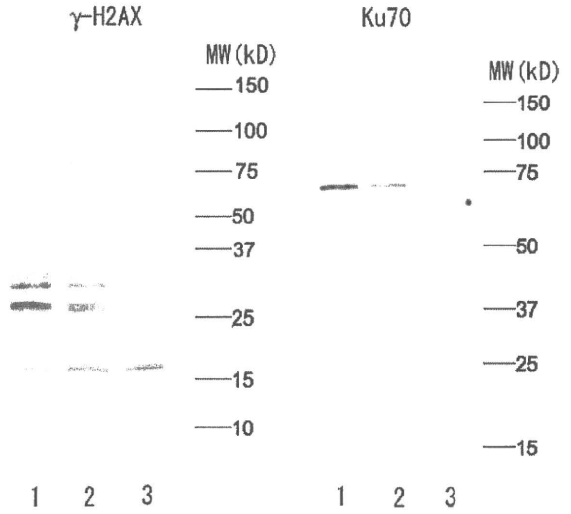

Fig. 2. Tests for the antibody specificity in immunoblotting. Muscle homogenates in control patients were segregated through polyacrylamide gel electrophoresis and immunoblotted using anti-γ-H2AX (left) and Ku70 (right) antibodies. The molecular weights of γ-H2AX and Ku70 are 15 kDa and 70 kDa, respectively. In γ-H2AX, patient 1 and 2, the extra bands between 25 kDa and 37 kDa correspond to the ubiquitinated forms [19]. In Ku70, patient 1 and 2, positive bands appear around its molecular weight.

ments, indicating nuclear breakdown. Ku70-positive deposits were sometimes found around intact nuclei (Fig. 3D).

Immunoblotting of muscle homogenates with the anti-Ku70 antibody showed a clear band around the molecular weight (Fig. 2).

3.3. Localization of HNE, iNOS, and LAMP-2

ROS is an inducer of DSB in muscle cells and oxidative stress may be associated with vacuolar formation [21], so we tested 4-hydroxy-2-noennal (HNE), a product of lipid peroxidation by ROS [22], and iNOS, a marker of oxidative stress that was previously found to be increased in vacuolated fibers [21]. HNE and iNOS were increased not only in some vacuolated fibers in s-IBM, but also in perifascicular atrophic fibers in dermatomyositis and ragged red fibers. In non-vacuolated fibers in s-IBM, atrophic fibers in neurogenic muscular atrophy, and pyknotic nuclear clumps, the two ROS markers were not increased.

Several studies indicated that rimmed vacuoles are lysosomes in origin [23,24]. In this study, we observed that vacuoles in s-IBM usually showed positive for the lysosome marker LAMP-2, as described previously [23]. A dual fluorescence study using antibodies against LAMP-2 and emerin showed frequent association of these two markers in the vacuoles in s-IBM.

3.4. Immuno-electron microscopy of Ku70

In the ultrastructural study of Ku70 in s-IBM, we detected Ku70-positive granules in some nuclei. Ku70-positive granules were often found in the vacuolar spaces of

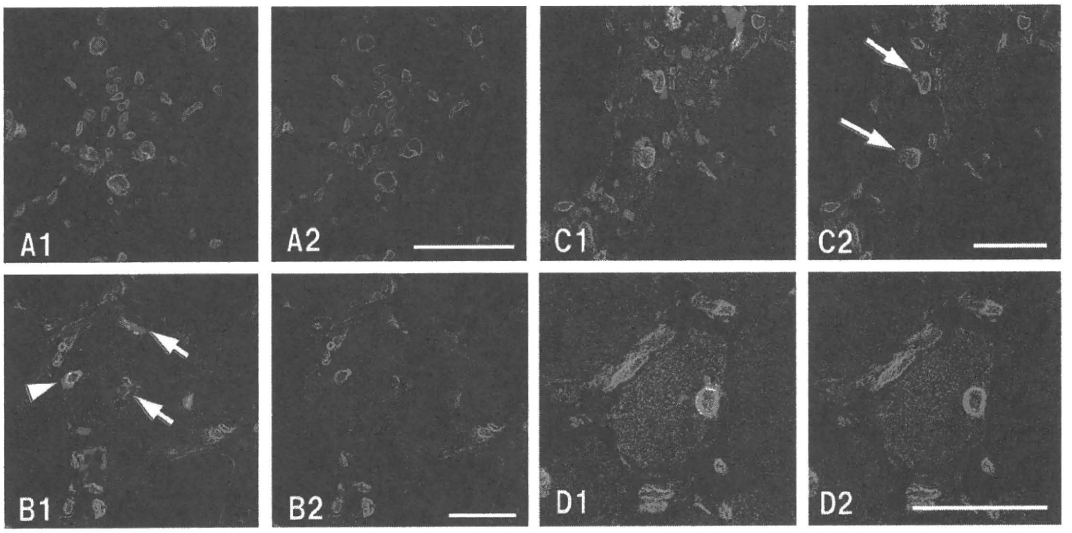

Fig. 3. Triple fluorescence study. Ku70 (red), emerin (green), and DNA (DAPI: blue). A: Regenerating fibers. B to D: s-IBM. A1–D1: overlay of the three colors. A2–D2: emerin plus DNA. (A) Ku70-positive deposits are largely confined to the area surrounded by emerin. (B) A vacuolated fiber contains a nucleus abutted by deposits of Ku70 (arrow head). Fragments of Ku70-positive deposits intermingle with remnants of emerin or DNA (arrows). The figures indicate nuclear breakdown and impaired incorporation of Ku70 into the nucleus. (C) Muscle fibers with numerous cytoplasmic deposits of Ku70 and breaks in the nuclear envelope (arrows). (D) Ku70-positive deposits surrounding a nucleus with an intact circle of emerin. This figure shows that nuclear import of Ku70 is impaired even in the early phase of nuclear breakdown in s-IBM. Bar = 25 μm.

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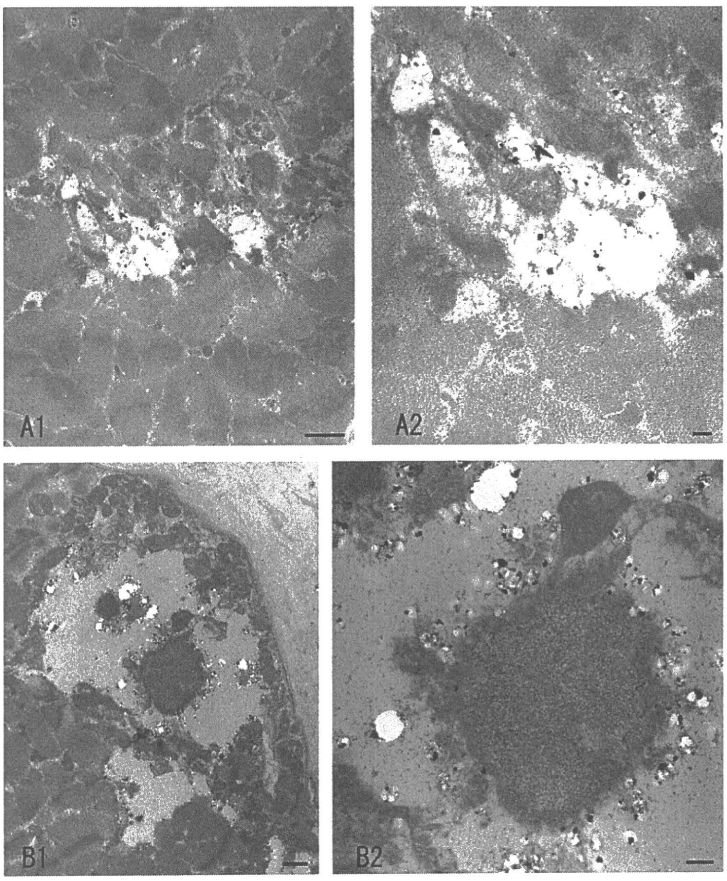

Fig. 4. Immunoelectron microscopy of Ku70, a regulatory component of DNA-PK. (A1) Deposits of Ku70 in cytoplasmic spaces. (A2) Higher magnification of A1. The positive reactivity may correspond to the cytoplasmic inclusion of Ku70 in immunofluorescence. (B1) Ku70 surrounding electron-dense round bodies. (B2) Higher magnification of one body shows granular structures inside with a peripheral dense zone. According to the triple fluorescence results, such as those shown in Fig. 2, the round body corresponds to a degenerating nucleus, and the electron micrograph may illustrate that the nucleus cannot incorporate Ku70. Bar = 1 μm (A1 & B1), 200 nm (A2), 500 nm (B2).

various sizes, sometimes combined with degenerative products (Fig. 4A). Ku70-positive products were also found to be attached to something like degenerating nuclear structures that contained no Ku70 (Fig. 3B), which may have corresponded to nuclei surrounded by Ku70-positive deposits in the immunofluorescence study (Fig. 2B, arrowhead).

4. Discussion

4.1. Findings in s-IBM

In the current study, we showed that the percentage of DSB-positive nuclei was significantly higher in vacuolated fibers than in other fibers in s-IBM. This finding suggests that nuclear breakdown along with the accumulation of DSB occurs in muscle cells in s-IBM. Moreover, we detected figures suggesting impaired nuclear import of Ku70. Nuclear translocation of Ku proteins is important for DBS repair, and a deficiency in nuclear translocation caused hypersensitivity against X-ray irradiation due to the lack of DBS repair in a cell culture study [25]. Therefore, we hypothesize that defects in Ku70 nuclear import accelerate DSB formation.

As DSB occur in other disease conditions without nuclear breakdown, additional factors may be involved in the nuclear changes seen in s-IBM. There is evidence that nuclear envelope dysfunction can cause both mechanical fragility of the nucleus and DNA damage. Lamins are proteins of nuclear intermediate filaments that comprise the lamina, the meshwork supporting inner nuclear membranes. Mutations in the genes that encode lamins and emerin (a lamin-associated protein) cause Emery–Dreifuss muscular dystrophy and a number of different diseases collectively called laminopathies [26]. In several laminopathies, blebbing of the nuclei in cultured fibroblasts can be seen, and it is hypothesized that such mutations result in fragile and mechanically unstable nuclei [27]. Emerin mutations can cause myopathy with rimmed vacuoles [28,29]. Besides structural integrity, the lamina is also involved in

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various other processes, such as replication and gene transcription, which are intimately associated with DNA damage repair. Accordingly, impaired DNA repair has been found in several laminopathies. Fibroblasts possessing a laminopathy mutation show an excessive amount of unrepaired DNA damage, as evidenced by γ-H2AX immunohistochemistry [30]. Furthermore, lamins are important in the spatial rearrangement of nuclear pore complexes and therefore nuclear protein transport. Nuclear protein import is reduced in cells expressing lamin A mutants [31]. In the current study, we detected figures suggestive of impaired nuclear import of Ku70. Defects of nuclear import have been suggested for the mechanism of cytoplasmic accumulation of enzymes (e.g., ERK [32] and MKP-1 [33]) and nuclear molecules (e.g., pElk-1 [5,32] and TDP-43 [34]) in s-IBM. In summary, dysfunctional lamins can explain the nuclear breakdown, accumulation of DSB, and impaired nuclear transport observed in s-IBM. A specific stressor predicted in this disease [35] may affect lamins or other nuclear envelope components. Alternatively, the nuclear envelope might become fragile by aging. Cell nuclei from old individuals exhibit defects similar to those of cells from Hutchinson–Gilford progeria syndrome, which is caused by mutations of lamin A [36]. Likewise, nuclear pore complexes are not turned over in differentiated cells, and age-related alterations in nuclear pore complexes affect nuclear integrity [37]. Moreover, several studies have indicated an age-dependent decline in DNA repair capacity [38]. We suspect that these age-associated changes in nuclear envelope integrity and DNA repair mechanisms may predispose the muscles of the elderly to s-IBM pathology. In this context, the initial inducer of DSB in s-IBM muscle may be the same as that in polymyositis.

We found products that were positive for the lysosome marker LAMP-2 in rimmed vacuoles, indicating that they also originate from lysosomes. Moreover, we found that the LAMP-2-positive products were frequently associated with emerin. These findings suggest the induction of autophagy to process broken-down nuclei. In the muscle of laminopathy patients and emerin-null mice, it has been shown that autophagosomes/autolysosomes are involved in the degradation of damaged nuclear components [39].

4.2. Findings in other diseases

Recent studies indicate an up-regulation of type 1 interferon inducible proteins in dermatomyositis muscle with perifascicular atrophy [40]. A prolonged stimulation of type 1 interferon (β-interferon) induces ROS and DNA damage response in culture study [41]. Therefore, the strong myonuclear γ-H2AX staining and excessive levels of ROS found in perifascicular atrophy might correspond with this hypothesis. The strong myonuclear γ-H2AX staining found in ragged red fibers may have been induced by increased ROS caused by mitochondrial dysfunction. Although the small angulated fibers show positive γ-H2AX reactivity, the majority of these fibers were negative for HNE and iNOS. Our results suggest that γ-H2AX histochemistry is more sensitive to detect ROS injury than the two markers or that other genotoxic stresses attack on muscle cells during degeneration. Contrary to the case in s-IBM, vacuoles in OPMD, which may originate from the nucleus and show some histone H1-positivity [5], were negative for γ-H2AX. This result suggests that simple nuclear breakdown does not induce DSB. We found a strong γ-H2AX reaction in a proportion of cells in inflammatory exudates. As DSB occurs during V(D)J recombination in lymphocyte development [11], γ-H2AX-positive cells may be active in gene recombination.

Acknowledgments

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ミオパチー

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■診断基準

ミオパチーとは筋疾患を意味する用語であり，何らかの理由で骨格筋が直接侵される疾患を指す．個々の筋疾患についての診断基準は存在するが，ミオパチー一般の診断基準は存在しない．

本稿では暫定的にミオパチーの診断基準として

① 筋力低下・筋萎縮・筋痙攣または血清クレアチンキナーゼ(CK)異常を呈する．

② 神経系または骨格系の異常を除外できる．

の2項目をあげたい．すなわちこれらの項目を満たした場合に，何らかの筋疾患が存在する可能性が高い．

■病型分類

ミオパチーの原因は遺伝性・感染性・代謝性・薬剤性に加えて原因不明までさまざまである．今回われわれは，過去の国内および海外での分類をもとに便宜的にTable 1のように病型分類する．以下，各病型について概説する．

1．筋ジストロフィー

筋ジストロフィーは一般的には「骨格筋の変性・壊死を主病変とし，臨床的には進行性の筋力低下をみる遺伝性の疾患である」と定義されている[1]．Duchenne/Becker型・Emery-Dreifuss型，肢帯型(LGMD)・先天性(福山型・非福山型)，顔面肩甲上腕型(FSHD)・眼咽頭型等に分類される．

Duchenne/Becker型は筋ジストロフィー全体のうち約50%を占め，四肢筋力低下による歩行障害に加え心不全など多彩な合併症があることから広く知られている．

2．筋強直症候群

筋強直症候群のうちもっとも有病率が高いのが，筋強直性ジストロフィーであり*DMPK*(dystrophia myotonica protein kinase)遺伝子の3′-非翻訳領域のCTG反復配列の伸張により発症する．筋強直性ジストロフィー2型(proximal myotonic myopathy：PROMM)は*ZNF-9*(zinc finger protein-9)遺伝子のイントロン領域のCCTG反復配列により発症する．臨床症状はきわめて多彩で，骨格筋症状(筋強直・心筋障害)のみならず，中枢神経(認知機能障害など)・内分泌(耐糖能機能異常など)および眼症状(白内障・網膜色素変性症)を合併する．

その他，先天性筋強直性ジストロフィー・先天性筋強直症(Thomsen病)・先天性パラミオトニアも筋強直症状を呈する．

3．遠位型ミオパチー

筋疾患の多くは体幹に近い四肢筋を侵すことが多いが，例外的に遠位筋優位に侵される一連の遺伝性筋疾患を遠位型ミオパチーという．神経原性筋疾患との鑑別が重要である．本邦では三好型ミオパチー・縁取り空胞を伴う遠位型ミオパチー(DMRV)・眼咽頭遠位型ミオパチーの3疾患が広く知られているが，その他Welander型遠位型ミオパチー(40代以降に発症し，手指伸筋の筋力低下を示す)・tibial muscular dystrophy(TMD)(前脛

Table 1. ミオパチーの病型分類

1．筋ジストロフィー Duchenne/Becker 型筋ジストロフィー Emery-Dreifuss 型筋ジストロフィー 肢帯型筋ジストロフィー 先天性筋ジストロフィー 顔面肩甲上腕型筋ジストロフィーなど	MERRF(ragged-red fiber を伴うミオクローヌスてんかん) CPEO(慢性進行性外眼筋麻痺)など
2．筋強直症候群 筋強直性ジストロフィー 先天性筋強直性ジストロフィー 先天性筋強直症(Thomsen 病) 先天性パラミオトニアなど	6．炎症性筋疾患 多発筋炎 皮膚筋炎 封入体筋炎など
3．遠位型ミオパチー 三好型ミオパチー 縁取り空胞を伴う遠位型ミオパチー 眼咽頭遠位型ミオパチー Welander 型遠位型ミオパチーなど	7．代謝性疾患 糖原病 VLCAD 欠損症 原発性カルニチン欠損症 多種アシル CoA 脱水素酵素欠損症など
4．先天性ミオパチー ネマリンミオパチー セントラルコア病 ミオチュブラーミオパチー 先天性筋線維タイプ不均等症など	8．内分泌性疾患 甲状腺ホルモン異常 副甲状腺ホルモン異常 副腎皮質ホルモン異常など
5．ミトコンドリア病 MELAS(高乳酸血症・卒中様症状を伴うミトコンドリア病)	9．中毒・感染性筋疾患 アルコール性ミオパチー ステロイドミオパチー スタチンミオパチー ウイルス・寄生虫感染症など
	10．周期性四肢麻痺

骨筋の筋力低下を示す)・distal *VCP*(valosin containing protein)-mutated myopathy(高齢発症で Paget 病と前頭側頭型認知症を伴う)・Miyoshi-like myopathy(三好型ミオパチーに臨床的に類似するが, dysferlin は正常)・筋原線維性ミオパチー(myofibrillar myopathy)などがある．筋原線維性ミオパチーは，病理学的な分類であり症候は多岐にわたっている．

4．先天性ミオパチー

臨床的には，①フロッピーインファント(floppy infant：生下時または乳児期早期より筋緊張低下を示す)，②頸部屈筋群の筋力低下，③顔面筋罹患(高口蓋)，④呼吸筋麻痺を呈することが多い．これらの症状に加え特徴的な筋病理所見を呈する疾患群を先天性ミオパチーと分類する．臨床経過からは生下時より呼吸障害や嚥下障害が強い重症型(severe infantile form)，発達遅延があるものの非進行性または緩徐進行性の良性先天型(benign congenital form)および成人発症型(adult onset form)の 3 型に分類される．診断は筋病理所見に基づいてなされる．ネマリンミオパチー・セントラルコア病・ミオチュブラーミオパチー・先天性筋線維タイプ不均等症(congenital fiber type disproportion：CFTD)などがある．

5．ミトコンドリア病

ミトコンドリア病はミトコンドリアの機能障害により何らかの症状を呈する病態であると定義できるが，通常は呼吸鎖酵素におけるエネルギー産生障害による疾患を指す．呼吸鎖は 5 種類の酵素複合体から構成される．酵素複合体に含まれる蛋白質はミトコンドリア DNA によりコードされるものと核 DNA によりコードされるものがある．いずれの異常によってもミトコンドリア病をきたしうる．ミトコンドリア DNA 変異は母系遺伝を

示すことがしばしばあるのに対し，核 DNA 異常は通常常染色体性遺伝を示す．

ミトコンドリア病は Table 1 に示した 3 大病型のように臨床症状により分類される場合と，生化学的な機能異常(ピルビン酸脱水素酵素複合体：PDHC 欠損症など)により分類される場合とがある．

6．炎症性筋疾患

多発筋炎・皮膚筋炎および封入体筋炎などが含まれる．

炎症性筋疾患の診断には臨床症状に加え，筋病理組織による診断が重要である．多発筋炎では，筋線維周囲および筋線維内へのリンパ球の浸潤がみられる．しばしば筋周鞘がアルカリホスファターゼ(alkaline phosphatase)染色で陽性を示す．皮膚筋炎では筋束周辺の筋線維萎縮(perifascicular atrophy)が特徴的変化として知られている．

成人の皮膚筋炎では，腫瘍性病変の合併頻度が高い．近年さまざまな自己抗体が同定されてきており，それぞれの抗体と臨床・病理学的変化の関連について整理されつつある．

封入体筋炎は高齢者にみられる疾患であり，大腿四頭筋と深指屈筋が高頻度に侵される．病理学的には，筋線維周囲へのリンパ球浸潤に加えて縁取り空胞を認めることが特徴である．

7．代謝性疾患

筋線維内でのエネルギー産生は，前述のミトコンドリアでの呼吸鎖のほかに細胞質内での解糖系や脂肪酸β酸化などに依存している．解糖系の異常で糖原病をきたす．とくに糖原病のうち，糖原病Ⅱ型(Pompe 病)では酸性α-グルコシダーゼ欠損によりライソゾーム内にグリコーゲンが蓄積し，肝細胞障害・肥大型心筋症・筋力低下を示す．近年，酵素補充療法が保険収載され，早期診断の重要性が増している．脂質代謝異常には VLCAD (very long chain acyl-CoA dehydrogenase)欠損症など横紋筋融解症をきたす疾患群のほか，原発性カルニチン欠損症や多種アシル CoA 脱水素酵素欠損症など脂質蓄積性ミオパチーをきたす疾患群がある．

8．内分泌性疾患

甲状腺ホルモン異常によりしばしば筋障害をきたす．甲状腺機能低下症では近位筋優位の筋力低下と易疲労性，緩徐な腱反射がしばしばみられる．またハンマーによる筋叩打により筋膨隆現象(mounding phenomenon)がみられる．血清 CK 値は無症候性でも高値を示すことがある．甲状腺中毒性ミオパチーは初期は近位筋の筋力低下を示すことが多く，ときに呼吸筋障害を呈することがある．血清 CK 値は正常または低下することがある．また甲状腺機能低下症・亢進症ともに重症筋無力症の合併頻度が高いことが知られている．

原発性アルドステロン症では低カリウム血症をきたしてミオパチーを呈する．後述の周期性四肢麻痺を参照されたい．

9．中毒・感染性疾患

中毒性疾患ではアルコール性ミオパチーやステロイドミオパチー，スタチンミオパチーなどの頻度が高い．アルコール性ミオパチーは急性壊死性ミオパチー・急性低カリウム性ミオパチー・慢性アルコール性ミオパチー・アルコール性心筋症といったさまざまな病型を含む．ステロイドミオパチーは下肢近位筋および下肢帯の筋力低下と筋萎縮を呈することが多い．血清 CK は正常または軽度の上昇にとどまる．スタチンミオパチーはスタチン(HMG-CoA 還元酵素阻害薬)の内服により誘発される横紋筋融解症を呈する．腎不全の合併が増悪因子となる．

感染性疾患は，主にウイルス性が多いがごくまれに寄生虫感染も報告される．ウイルス性筋炎の原因として，コクサッキー・エコー・インフルエンザ・HIV・パルボウイルスなどが知られている．

10．周期性四肢麻痺

発作性に四肢および体幹の弛緩性麻痺を呈する．内分泌異常を除外したものを原発性周期性四肢麻痺という．イオンチャネルをコードしている遺伝子変異により筋線維の膜電位異常が起こると

考えられる．発作中の血清カリウムイオン濃度により高・低カリウム性周期性四肢麻痺に分けられる．

低カリウム性周期性四肢麻痺の原因として*CACNA1S*(calcium channel, voltage-dependent, L type, alpha-1S subunit)遺伝子の異常が同定されている．高カリウム性周期性四肢麻痺の原因としては*SCN4A*(sodium channel, voltage-gated, type Ⅳ, alpha subunit)遺伝子の異常が同定されている．筋強直症候群の項であげた先天性パラミオトニアは同一の遺伝子異常による allelic な疾患である．

■重　症　度

筋力自体の評価に加えて，筋力低下による歩行障害などの日常生活動作(activities of daily living：ADL)制限・嚥下機能障害による摂食量の低下や誤嚥・呼吸筋力低下による換気障害・心筋機能低下等の評価が重要である．

筋力は，一般に徒手筋力検査(manual muscle testing：MMT)で評価される．ただし，検者間での差異があるため，ダイナモメーターやピンチメーターによる定量的評価を行うこともある．握力は MMT に比較して客観的であると考えられるが，器材による違いがある．また，加速度計による運動の評価も行われる．血清 CK 値は通常，筋線維の壊死を反映するが，進行期ではむしろ低下することに注意が必要である．CK 値の増加がみられない筋疾患もある．ミトコンドリア病では通常血清および髄液中の乳酸の上昇がみられる．筋量の評価には骨格筋 CT が用いられる．炎症性筋疾患では，炎症部位の同定に筋 MRI が有用である．

嚥下機能評価にはしばしば食道嚥下造影検査が用いられる．嚥下性肺炎の危険性評価に加え，経口栄養から経管栄養への切り替えを判断する際に有用である．

肺機能検査や動脈血ガス分析により呼吸筋障害の程度を評価する．

心臓超音波検査で心拍出量や心筋の運動量を調べて心筋障害の程度を評価する．血液生化学的検査では脳性ナトリウム利尿ペプチド(brain natriuretic peptide：BNP)が心不全の指標として有用である．

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6 筋疾患とオートファジー

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疾病研究第一部

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2008年3月東京理科大学院基礎工学研究科生物工学専攻卒業，同年4月国立精神・神経センター神経研究所疾病研究第一部流動研究員。研究テーマは，ダノン病，XMEA治療法開発に向けた分子病態の解明。趣味は，サッカー。

Key words：ミオパチー，オートファジー，自己貪食空胞，遺伝性筋疾患

Abstract
筋細胞におけるオートファジーの役割，およびリソソームの機能は長らく不明であった。しかし，自己貪食空胞が蓄積する筋疾患の存在から，筋細胞におけるリソソームとオートファジーの重要性が注目されるようになっている。オートファジーの異常を示す筋疾患は，その原因遺伝子と病理観察から，①オートファジー/リソソーム系の異常によるものと，②オートファジーそのものの異常ではなく，2次的にオートファジーが惹起されるものの2つに分類することができる。

1. 骨格筋・心筋におけるオートファジー研究

正常な骨格筋・心筋においては，形態学的観察でリソソームや自己貪食空胞が認められることは少なく，骨格筋・心筋におけるオートファジーの役割，およびリソソームの機能は長らく不明のままであった。しかしながら，自己貪食空胞が蓄積する筋疾患の存在が明らかになり，筋肉におけるリソソームとオートファジーの重要性が重視されるようになってきた。我々はこれまでにオートファジーの機能異常により筋線維内に自己貪食空胞が蓄積する自己貪食空胞性ミオパチー(Autophagic vacuolar myopathy：AVM)という一群の筋疾患を提唱し，広く世界に受け入れられている。

原因遺伝子の明らかになっているAVMとして，ダノン病・X-linked myopathy with excessive autophagy (XMEA)・酸マルターゼ欠損症・縁取り空胞型遠位型ミオパチー(distal myopathy with rimmed vacuoles：DMRV)があげられる。それぞれの原因遺伝子はLAMP-2 (lysosome-associated membrane protein-2)・Vma21・酸マルターゼ・GNE（ウリジン二リン酸-N-アセチルグルコサミン2-エピメラーゼ／N-アセチルマンノサミンキナーゼ）である。LAMP-2はリソソーム膜タンパク質，Vma21はV-ATPaseの複合体形成因子，酸マルターゼはグルコース分解を行うリソソーム酵素，GNEはシアル酸合成に必須な2つの酵素をコードしている。

このようにAVMは原因遺伝子から，①オートファジー / リソソーム系そのものの機能異常により病態を示すもの，②オートファジー / リソソーム系の機能異常が本質的な疾患原因ではなく，2次的にオートファジーの働きが惹起し病態を示すものの2つに分類される。前者ではダノン病・XMEA・酸マルターゼ欠損症，後者ではDMRVが代表的な筋疾患として分類される。以下，それぞれの筋疾患に対しての病理症状およびこれまでの研究成果について記す。

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2. 自己貪食空胞性ミオパチー（AVM）

1）ダノン病

ダノン病は，X連鎖優性遺伝のまれな疾患で，全世界で数十例の報告があるにすぎない。ダノン病は，X染色体上にコードされているリソソームの膜タンパク質の1つLAMP-2が欠損することにより起こる遺伝性疾患であり，精神遅滞・ミオパチー・肥大型心筋症を3徴とする。ダノン病は症状が潜行性のため早期の発見が難しいが，すべての患者で進行性の心筋症がみられる。この心筋症はしばしば不静脈を伴い突然死にいたることがあり，現在のところ心臓移植以外に効果的な治療法は報告されていない。患者の筋組織をみると，筋線維内に多数の小胞が蓄積しており，電子顕微鏡観察から，それらが自己貪食空胞であることが分かる。また，これら蓄積空胞の周囲には，それを取り囲むように筋鞘膜様の特徴をもつ膜 (autophagic vacuoles with sarcolemmal features：AVSF)が観察される（図1-A)。AVSFでは空胞膜にほぼすべての筋鞘膜タンパク質が発現しており，アセチルコリンエステラーゼ活性を有している。これら特徴からAVSF形成は，細胞が自身の内部に細胞外環境を作り出しているかのような状況であるといえる。しかしながら，現在のところ，このAVSFがどのようにして形成されるのか？病態への関与は何なのか？などといった疑問は解明されておらず，そもそもAVSF形成が，生体防御的に働いているのか，あるいは病態悪化要因であるのかさえ不明である。

LAMP-2は，その常染色体上ホモログであるLAMP-1と共に，リソソーム膜の約50%を構成するリソソーム膜主要構成タンパク質であり，ノックアウトマウスを用いた研究からリソソームの移動・オートファゴソームとの融合に関与していることが明らかになっている。LAMP-2は正常では心臓・骨格筋・脳において強く発現しており，ダノン病では，これらの領域において，恒常的におこっているオートファジーがその最終段階（オートファゴソームとリソソームの融合段階）でストップしており，症状を呈するようになると考えられる。

2）X-linked myopathy with excessive autophagy (XMEA)

XMEAはX染色体劣性遺伝形質をとる遺伝性筋疾患であり，女性保因者は発症しない。この疾患は臨床的にはダノン病よりも軽く，心筋障害もきたさないが，筋組織所見は極めて類似している。近位筋優位の進行性の筋力低下と委縮が認められるが，程度は軽く，60歳を過ぎても歩行可能な例が多い。

XMEAは長らくその原因遺伝子が不明のままであったが，2009年についにその原因遺伝子がVMA21であることが明らかになった。VMA21はV-ATPaseの複合体構成因子であり，その欠損はV-ATPaseの活性を低下させる。V-

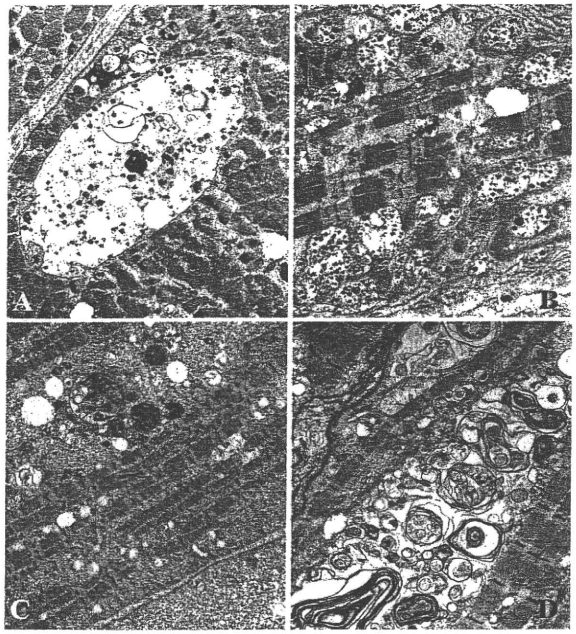

図1　AVM骨格筋の電子顕微鏡写真
A：ダノン病，B：XMEA，C：酸マルターゼ欠損症，D：DMRV

ATPaseはリソソーム内を酸性に保つためのプロトンポンプとして働いており，その機能低下によりリソソーム内のpHが上昇し，タンパク質の分解が低下する。つまり，XMEAではVMA21の変異により，リソソームの分解能が低下することで，オートファジーの機能異常がおこり病状を呈する。

XMEAの筋病理ではダノン病と同様に筋線維内にAVSFを認める。通常AVMの電子顕微鏡観察では，蓄積した自己貪食空胞内には分解前の細胞内小器官，分解された不定形の分解物のどちらかを認めるが，XMEAにおいては空胞内に電子密度の高い円い顆粒状の内容物が観察される（図1-B）。さらに，蓄積された空胞があたかもエキソサイトーシスにより細胞外に放出されているような像も観察される。また筋鞘膜部の基底膜が肥厚化し，自己貪食空胞内に見られたものと同様の細胞質分解産物の蓄積が観察される。これらの現象の意義はいまだ明らかになっていないが，この現象は，細胞が分解できずに蓄積した空胞を，エキソサイトーシスの経路に乗せかえることにより，細胞外に放出している可能性を推測させる。

3）酸マルターゼ欠損症

酸マルターゼ欠損症は糖原病II型として分類される常染色体劣性遺伝性の疾患である。酸マルターゼはグリコーゲン代謝に必須のリソソーム酵素であり，その機能不全によりリソソーム内外に未分解のグリコーゲンが異常に蓄積する（図1-C）。

酸マルターゼ欠損症は，臨床的に乳児型・小児型・成人型の3つに分類される。臨床的に最も重篤な症状を示す乳児型は，出生時より筋力の低下，筋緊張の低下と肥大型心筋症を呈し，心肺機能不全により2歳以前に死亡する。小児型は幼児期あるいは幼年期に呼吸筋の筋力低下として発症するが，乳児型とは異なり，通常，心臓の障害は認められない。また成人型は主として20歳以降に発症し，肢体筋の筋力低下・萎縮を伴う。小児型と同様に呼吸筋の低下が見受けられ，呼吸不全を初期症状とする。小児型と成人型はまとめて遅発型と呼ばれることもある。酸マルターゼ欠損による筋障害発症機構は明らかにはなっていないが，現在のところ,その機能不全によりリソソーム内さらには2次的に細胞質にグリコーゲンが蓄積し，それによりオートファジーが盛んにおこることで,筋線維を破壊し，筋障害にいたるのではないかと考えられている。

4）縁取り空胞型遠位型ミオパチー(DMRV)

DMRVは，第9染色体にある*GNE*遺伝子の主にミスセンス変異を原因とする常染色体劣性遺伝性疾患である。*GNE*はシアル酸生合成経路の律速酵素であり，また，高等生物ではこれが唯一のシアル酸合成経路であるため，*Gne*欠損マウスは胎生致死を示す。

DMRVは1981年に埜中らにより報告されたもので，遺伝性封入体ミオパチーもしくは，埜中ミオパチーとも呼ばれている。筋生検をすると，筋線維の中に細かい顆粒状の物質で縁取られた空胞(縁取り空胞：rimmed vacuole）が検出されるのが特徴的である。患者は10代後半～30代後半に掛けて発症し，主に，遠位筋の筋力低下と筋萎縮をきたし，10年程度で急速に歩行不能となる進行性の筋疾患である。先にも述べたように，筋病理所見として縁取り空胞を認めるが，この縁取り空胞は実際には染色操作の過程でできる人工産物である。電子顕微鏡観察では，筋線維内の縁取り空胞部分には，堆積したもろい自己貪食空胞とミエロイド小体が蓄積していることが確認される（図1-D）。

DMRVは*GNE*の機能喪失型変異により発症する。これまでの研究から，低シアル酸状態が各種の病理学的変化を引き起こしていることが明らかとなっている。また，本疾患では，縁取り空胞内部あるいは周辺部にβ-アミロイドが蓄積することが明らかになっている。これらの結果から，何らかの理由により，本来は分解されて低レベルでしか存在しないはずの異常タンパク質や基質が蓄積し，結果的にオートファジーの機能が惹起しているものと推測される。

3. 治療法開発に向けて

他の遺伝性疾患同様に，AVMも現在のところ完全な治療法は確立されていない。しかしながら，酸マルターゼ欠損症およびDMRVに対して，今後の展開が期待される治療法が開発されつつある。

酸マルターゼ欠損症に対しては，アメリカのGenzyme社が開発した組み換え型ヒトα-グルコシダーゼによる酵素補充療法剤による治療がおこなわれており，乳児型では明らかな効果が得られている。しかしながら，問題点として，効果が長続きせず，2週間に1回の点滴静注が必要となることがあげられている。

また，我々の研究室では，DMRVのモデルマウスを世界で初めて作成し，そのマウスを用いた研究から，原因遺伝子*GNE*によりコードされる酵素の最終代謝産物であるシアル酸，シアリル乳糖または生合成中間体であるN-アセチルマンノサミンを投与することにより，病態の進行をほぼ完全に抑制することが可能であることを明らかにし，DMRVの治療に向け大きな1歩を進めることに成功した。

おわりに

筋細胞におけるオートファジーの研究は，AVM患者数の問題などからもいまだ十分になされているとは言い難い。しかしながら我々や他の研究者たちの成果により確実にその役割・重要性が明らかになりつつある。オートファジーの異常をしめす筋疾患は，オートファジー/リソソーム系の異常によるものと，オートファジーそのものの異常ではなく，2次的にオートファジーの機能が惹起されているものがあり，前者ではAVSF，後者では縁取り空胞が特徴的な病理症状としてあげられる。現在のところ，酸マルターゼ欠損症・DMRVに対しては，我々や他の研究者により治療の可能性が示されているものの，他のAVMに対する効果的な治療法は開発されておらず，今後のより詳細な病態解明および，それに基づいた治療法の開発が期待される。また，AVMにおける自己貪食空胞の蓄積メカニズムは十分に解明されたとは言いがたく，その詳細を解明することは，正常組織でのオートファジーの役割を理解する上で重要であると考えられる。

平成22年度　班会議プログラム

厚生労働科学研究費補助金　難治性疾患克服研究事業

封入体筋炎（IBM）の臨床病理学的調査および診断基準の精度向上に関する研究班

平成２２年度　研究班会議プログラム

研究代表者　　東北大学病院　神経内科

青木　正志

日　　時　　平成 23 年 1 月 29 日(土) 14:00～17:10

会　　場　　東北大学病院　東病棟 4 階　第五会議室

〒980-8574 宮城県仙台市青葉区星陵町 1-2

ＴＥＬ　022-717-3792 (会議室) / 7189 (医局)

お願い：演題発表時間 15 分（発表 10 分、討論 5 分）
発表者はご自身の PC をお持ちいただきますようお願いいたします。

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厚生労働科学研究費補助金　難治性疾患克服研究事業
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開会挨拶および本研究班の主旨について　　14:00～14:10　研究代表者　青木 正志

封入体筋炎に関する班員研究発表

Session I　　14:10～14:55　座 長　内野 誠

1. 重篤な嚥下障害を来した抗 SRP/抗 RNP 抗体陽性壊死性ミオパチーの 2 症例

研究分担者　森 まどか 1

共同研究者：○池田謙輔 1、山本敏之 1、大矢　寧 1、西野一三 2、村田美穂 1

所属：1 国立精神・神経医療研究センター病院　神経内科
2 国立精神・神経医療研究センター　神経研究所　疾病研究第一部

2. 封入体筋炎における ALS 関連分子の筋病理学的検討

研究分担者　内野　誠

共同研究者：○山下　賢、木村　円、俵　望、坂口秀哉、中間達也、前田　寧、平野照之

所属：熊本大学神経内科

3. 封入体筋炎における筋エコーの有用性について

研究分担者：梶 龍兒

共同研究者：○松井尚子，鎌田えりか，高松直子，寺澤由佳，和泉唯信

所属：徳島大学神経内科

Session II　　14:55～15:40　座 長　西野 一三

4. 嚥下障害を有する封入体筋炎患者へのバルーンカテーテル拡張法の検討

研究分担者　近藤 智善

共同研究者：○村田 顕也

所属：和歌山県立医科大学神経内科