Vacuolar myopathies

Vacuolar myopathies comprise a heterogeneous group of disorders which share as their main histopathological feature the presence of vacuoles. A vacuole is defined as an abnormal space within the muscle fibre that appears empty by at least one method of staining (Engel AG 1973a) (1). In general vacuoles may be divided into two categories: (a) membrane bound and (b) non-membrane bound, also described as a cytoplasmic space.

A vacuole may contain an excess of normal material, such as glycogen and/or lipid, or an excess of abnormal material such as cytoplasmic degradation products. Although usually no one vacuole is specific for any one disease, certain types of vacuoles tend to be associated with a specific disorder. Electron microscopy provides important information on the characteristics and contents of the vacuoles, which in turn leads to pattern recognition of some of the more common disease entities (Chapter KK, TK) (2).

Lysosomal myopathies is an area in which electron microscopy plays an important diagnostic role. These may be defined as myopathies due to primary or secondary lysosomal defects and are characterized morphologically by the presence of vacuoles called autophagosomes. There are three main categories of lysosomal myopathies;

1. Acid maltase deficiency
2. Myopathies characterized by rimmed vacuoles
3. Myopathies with autophagic vacuoles with unique sarcolemmal features

Among the myopathies with autophagic vacuoles with unique sarcolemmal features there are five recognized myopathies although two and five (see below) may be allelic. They are all characterized by the presence of vacuoles, whose membranes exhibit sarcolemmal features including the presence of sarcolemmal specific proteins such as dystrophin, sarcoglycans, dystroglycans, laminin and acetyl cholinesterase.

1. Danon disease – originally called lysosomal glycogen storage disease with normal acid maltase (3,4). This is the only myopathy where the gene defect is known and is due to mutations in the lysosome-associated membrane protein-2 (LAMP-2).
2. X-linked myopathy with excessive autophagy (XMEA) (5).
3. Infantile autophagic vacuolar myopathy (6).
4. Adult onset autophagic vacuolar myopathy (7,8).
5. X-linked congenital autophagic vacuolar myopathy (9).

Discussion

Of the above categories the underlying molecular defect has been characterized only in Danon’s disease; this involves mutations in the LAMP-2 gene which usually cause loss of
expression of the LAMP-2 protein. In our case the subsarcolemmal vacuoles were more prominent and intracytoplasmic vacuoles were rare. Cytoplasmic vacuoles showed positive staining for spectrin, dystrophin, laminin and dystrolycans. In addition positive staining was seen for acetylcholinesterase, C5b9 and LAMP-2 antibodies. The vacuoles were evident with H+E staining and contained basophilic material; they were also positive for acid phosphatase and non-specific esterase. H+E stained sections also demonstrated a substantial variability in fibre size and the presence of endomysial fibrosis which was also evident in Gomori trichrome. Acid maltase was normal in our case, although some of the intracytoplasmic vacuoles contained glycogen.

By electron microscopy the vacuolar membranes had basal lamina on the luminal side of the membrane which confirmed that vacuolar membranes had sarcolemmal features. Electron microscopy also showed the presence of electron dense myeloid type material within the subsarcolemmal vacuoles and there was reduplication of the basal lamina. Based on the light and electron microscopical results we believe that our case fulfills the criteria of an autophagic vacuolar myopathy with unique sarcolemmal features but does not fit clinically or histologically into any of the five categories already described above.

Overall the biopsy of this patient had ample expression of LAMP-2 and hence it can not be a case of Danon myopathy, furthermore the clinical features and the absence of cardiomyopathy are not typical of this disease. The presence of acetyl cholinesterase and C5b9 in the sarcolemma is typical of non-Danon lysosomal myopathies yet the distribution of the vacuoles which were in the great majority subsarcolemmal as well as the clinical features of the patient raise the possibility of further heterogeneity in this group of disorders.

Working diagnosis in our case

Autophagic vacuolar myopathy with unique sarcolemmal features? New type.

References

Slide 1. H+E showing presence of subsarcolemmal (arrowheads) as well as intracytoplasmic vacuoles (arrows). There is a marked variability in fibre size, increased endomysial fibrosis, necrosis and regeneration (x400).

Slide 2. Gomori trichrome showing presence of subsarcolemmal (arrowheads) as well as intracytoplasmic vacuoles (arrows) (x400).

Slide 3. ATPase pH9.4 showing type 1 fibre predominance, pale brown and round atrophy affecting both fibre types (x200).

Slide 4. Acid phosphatase staining showing increased punctate activity in some myofibres (arrows). A necrotic fibre is also seen (arrowheads) (x400).

Slide 5. Resin semithin section stained with toluidine blue, showing presence of numerous subsarcolemmal vacuoles (arrows) (x400).

Slide 6. Immunofluorescence staining for δ-sarcoglycan showing staining of subsarcolemmal vacuole (arrow) (x400).
Slide 7. Immunofluorescence staining for LAMP-2 showing staining of subsarcolemmal vacuoles (arrows) (x400).

Slide 8. Immunofluorescence staining for C5b-9 showing staining of subsarcolemmal vacuoles (arrows) (x400).

Slide 9. Electron micrograph showing the presence of subsarcolemmal vacuoles containing osmiophilic material (arrows). Note presence of basal lamina within the luminal side of the vacuole (arrowheads) (x40,000).

Slide 10. Electron micrograph showing the presence of an intracytoplasmic vacuole containing glycogen (arrows) (x70,000).