Muscular dystrophies involving the dystrophin–glycoprotein complex: an overview of current mouse models

Madeleine Durbeej and Kevin P Campbell*

The dystrophin–glycoprotein complex (DGC) is a multisubunit complex that connects the cytoskeleton of a muscle fiber to its surrounding extracellular matrix. Mutations in the DGC disrupt the complex and lead to muscular dystrophy. There are a few naturally occurring animal models of DGC-associated muscular dystrophy (e.g. the dystrophin-deficient *mdx* mouse, dystrophic golden retriever dog, HFMD cat and the δ-sarcoglycan-deficient BIO 14.6 cardiomyopathic hamster) that share common genetic protein abnormalities similar to those of the human disease. However, the naturally occurring animal models only partially resemble human disease. In addition, no naturally occurring mouse models associated with loss of other DGC components are available. This has encouraged the generation of genetically engineered mouse models for DGC-linked muscular dystrophy. Not only have analyses of these mice led to a significant improvement in our understanding of the pathogenetic mechanisms for the development of muscular dystrophy, but they will also be immensely valuable tools for the development of novel therapeutic approaches for these incapacitating diseases.

Addresses
Howard Hughes Medical Institute, Department of Physiology and Biophysics, Department of Neurology, University of Iowa College of Medicine, Iowa City, Iowa 52242, USA
*e-mail: kevin-campbell@uiowa.edu

Abbreviations

CMD	congenital muscular dystrophy
DGC	dystrophin–glycoprotein complex
DMD/BMD	Duchenne or Becker muscular dystrophy
FCMD	Fukuyama congenital muscular dystrophy
HFMD	hypertrophic feline muscular dystrophy
LGMD	limb–girdle muscular dystrophy
Mdx	X-chromosome-linked muscular dystrophy
MEB	muscle–eye–brain disease
myd	myodystrophy
nNOS	neuronal nitric oxide synthase

Introduction

Muscular dystrophy is a general term that describes a group of inherited and gradually debilitating myogenic disorders. Genetically, the pattern of inheritance can be X-linked recessive as in Duchenne or Becker muscular dystrophy (DMD/BMD), autosomal dominant as in limb–girdle muscular dystrophy type 1 (LGMD type 1), or autosomal recessive as in limb–girdle muscular dystrophy type 2 (LGMD type 2). DMD is the most common type of muscular dystrophy affecting approximately 1 out of 3500 males whereas the limb–girdle muscular dystrophies affect roughly 1 out of 20,000. Clinically, the muscular dystrophies are a heterogeneous group of disorders. Patients with DMD have a childhood onset phenotype and die by their early twenties as a result of either respiratory or cardiac failure, whereas patients with BMD have moderate weakness in adulthood and may have normal life spans. The limb–girdle muscular dystrophies have a highly variable onset and progression, but the unifying theme among the limb–girdle muscular dystrophies is the initial involvement of the shoulder and pelvic girdle muscles. Moreover, muscular dystrophies may or may not be associated with cardiomyopathy [1–4].

Combined positional cloning and candidate gene approaches have been used to identify an increasing number of genes that are mutated in various forms of muscular dystrophy. According to the genetic basis, muscular dystrophies have now been reclassified and close to 30 genes have been implicated to cause muscular dystrophy (see [5] for review; see also Table 1). The first gene to be cloned was the dystrophin gene that is mutated in DMD and BMD [6]. Soon after the discovery of dystrophin, the dystrophin–glycoprotein complex (DGC) was identified and these studies opened up a new avenue of muscular dystrophy research [7–9]. Within the past couple of years, several targeted mouse models for DGC-associated muscular dystrophy have been generated and these mouse models, which are the focus of this review, have significantly contributed to understanding the pathogenetic mechanisms of muscular dystrophy.

Dystrophin–glycoprotein complex

The DGC is a large complex of membrane-associated proteins that is critical for the integrity of skeletal muscle fibers. This complex consists of dystrophin, the dystroglycans (α and β), the sarcoglycans (α, β, γ and δ), sarcospan, the syntrophins (α1, β1, β2; γ1- and γ2-syntrophins have been identified in neurons) and α-dystrobrevin [7,8,10–21]. Dystrophin binds to cytoskeletal actin and to the transmembrane protein β-dystroglycan; the extracellular domain of β-dystroglycan binds to the peripheral membrane protein, α-dystroglycan; and α-dystroglycan binds laminin-2 in the basal lamina [22–24] (see Figure 1). Furthermore, α-dystroglycan has been shown to bind the heparane sulfate proteoglycans agrin and perlecan and to the chondroitin sulfate proteoglycan biglycan [25–28]. In addition, α-dystroglycan was recently shown to bind neurexin in neurons [29]. Perlecan and agrin are, along with laminin-2, also present in the basement membrane of the skeletal muscle. Thus, the DGC serves as a link between the extracellular matrix and the subsarcolemmal cytoskeleton and the DGC is thought to protect muscle cells from contraction-induced damage [30–31]. In agreement with this hypothesis, mutations in

Table 1

Muscular dystrophies and corresponding mouse models.

| Disease	Mode of inheritance and gene locus		Gene product	Mouse models
X-linked MD				
Duchenne/Becker MD	XR	Xp21	Dystrophin	Mdx
Emery–Dreifuss MD	XR	Xp28	Emerin	–
Limb–girdle MD				
LGMD 1A	AD	5q31	Myotilin	–
LGMD 1B	AD	1q11	Lamin A/C	Lmna$^{-/-}$ [121]
LGMD 1C	AD	3p25	Caveolin-3	Cav3$^{-/-}$
LGMD 1D	AD	6q23	?	–
LGMD 1E	AD	7q32	?	–
LGMD 1F	AD	5q31	?	–
LGMD 2A	AR	15q15	Calpain-3	Capn3$^{-/-}$ [122]
LGMD 2B	AR	2p13	Dysferlin	SJL [123]
LGMD 2C	AR	13q12	γ-sarcoglycan	Sgcg$^{-/-}$
LGMD 2D	AR	17q12	α-sarcoglycan	Sgca$^{-/-}$
LGMD 2E	AR	4q12	β-sarcoglycan	Sgcb$^{-/-}$
LGMD 2F	AR	5q33	δ-sarcoglycan	Sgcd$^{-/-}$
LGMD 2G	AR	17q11	Telethonin	–
LGMD 2H	AR	9q31	TRIM31 [124]	–
LGMD 2I	AR	19q13	Fukutin-related protein [125]	–
Distal MD				
Miyoshi myopathy	AR	2p13	Dysferlin	SJL [123]
Tibial MD	AD	2q31	?	–
Congenital MD				
Classical or pure CMD	AR	6q22	Laminin α2	dy
Fukuyama CMD	AR	9q31	Fukutin	–
MDC1C	AR	19q13	Fukutin-related protein [125]	–
α7 integrin CMD	AR	12q13	α7 integrin	Itga7$^{-/-}$
Ulrich CMD	AR	?	Collagen VI α2 [126]	–
Walker Warburg syndrome [127]	AR	?	?	–
Rigid spine CMD	AR	1p35	Selenoprotein N [128]	–
Muscle–eye–brain disease	AR	1p32	POMGnT1	–
Other forms of MD				
Emery-Dreifuss MD	AD	1q11	Lamin A/C	Lmna$^{-/-}$ [121]
Bethlem myopathy	AD	21q22	Collagen V1 α1	Col6α1$^{-/-}$ [129]
Bethlem myopathy	AD	21q22	Collagen V1 α2	–
Bethlem myopathy	AD	2q37	Collagen V1 α3	–
?	?	?	Collagen XV [130]	Col15α1$^{-/-}$
EB and MD	AR	8q24	Plectin	Plectin$^{-/-}$ [131]
Facioscapulohumeral MD	AD	4q35	?	–
Scapuloperoneal MD	AD	12q21	?	–
Oculopharyngeal MD	AD	14q11.2	Poly A binding protein 2	–
Myotonic dystrophy	AD	19q13	Myotonin-protein kinase/Six5	Six5$^{-/-}$ [132,133]

A summary list of muscular dystrophies, their Mendelian inheritance pattern, chromosomal location, mutated proteins and available mouse models. MD, muscular dystrophy; LGMD, limb–girdle muscular dystrophy; CMD, congenital muscular dystrophy; EB, epidermolysis bullosa; XR, X-linked recessive; AD, autosomal dominant; AR, autosomal recessive. For primary references on the muscular dystrophies, their mode of inheritance, gene locus, mutated proteins and available mouse models, please see [5] and main text.

genes encoding dystrophin, all four sarcoglycans and the laminin α2 chain are responsible for DMD/BMD, limb–girdle muscular dystrophy type 2C-F and congenital muscular dystrophy (CMD), respectively [6,11,14–17,32,33]. Not only does the DGC have structural roles but it may also play a role in signaling. Several signaling molecules bind DGC core components. Grb2, a signal transduction adapter protein, binds β-dystroglycan [34]. The filamins have been implicated as signal transducers and a member of the filamin family, filamin 2, interacts with γ- and δ-sarcoglycan [35]. Furthermore, dystrophin interacts with two classes of cytoplasmic molecules, the syntrophins [36,37] and dystrobrevins, which have

been implicated in signaling [21]. α1-syntrophin contains a PDZ-domain (a protein–protein interaction motif) that interacts with at least two sarcolemmal proteins involved in signal transduction, neuronal nitric oxide synthase (nNOS) [38] and voltage-gated sodium channels [39,40]. Similarly, the PDZ-domain of β2-syntrophin (expressed at the neuromuscular junction where it binds the dystrophin homologue utrophin) interacts with the PDZ-domain-containing microtubule-associated serine/threonine kinases MAST205 and SAST [41]. α-dystrobrevin binds syntrophins in addition to dystrophin and is a substrate for tyrosine kinases [42,43]. Moreover, biochemical evidence was recently presented for

Figure 1

The DGC in skeletal muscle is composed of dystrophin, the dystroglycans (α, β), the sarcoglycans (α, β, γ, δ), sarcospan, the syntrophins (α, β1) and dystrobrevin (α). There is a growing number of proteins reported to be associated with the DGC in the muscle cell. Depicted are nNOS, which interacts with the syntrophin complex and laminin-2, which is one of many extracellular ligands of α-dystroglycan. Several forms of muscular dystrophy arise from primary mutations in genes encoding components of the DGC. Mutations in dystrophin, all four sarcoglycans and the laminin α2 chain are responsible for DMD/BMD, LGMD type 2C-F and CMD, respectively. In addition, several forms of CMD are caused by abnormal glycosylation of α-dystroglycan (not illustrated).

Current Opinion in Genetics & Development
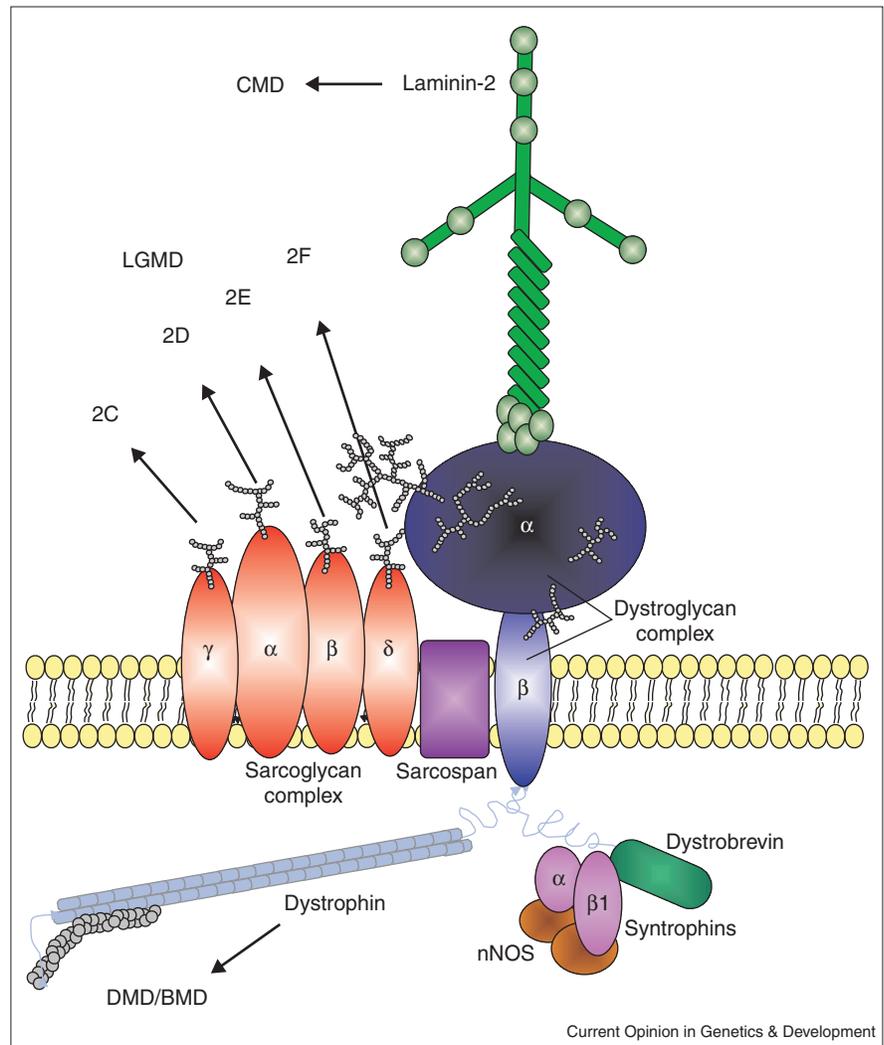
an association of α-dystrobrevin with the sarcoglycan–sarcospan complex, indicating that the sarcoglycan complex is linked to nNOS signaling via α-syntrophin/α-dystrobrevin [44].

All vertebrates seem to have well-defined sequences encoding dystrophin and its homologues, utrophin and DRP-2. Dystrophin-like sequences have also been identified in invertebrates such as amphioxus, sea squirt, starfish, scallop, fruit fly and nematode [45]. In each case, phylogenetic analyses showed the invertebrate dystrophins to be orthologues of the last common ancestor of dystrophin, utrophin and DRP-2. Given the diversity of the remainder of the components of the vertebrate DGC, how much simpler is the invertebrate counterpart? The release of the complete sequences of the *Drosophila melanogaster* and *Caenorhabditis elegans* genomes have afforded the opportunity to assess the invertebrate repertoire of the DGC, using a combination of *in vitro* and *in silico* techniques [46,47] (Table 2). In summary, seventeen known human/mouse components (three dystrophin-related proteins, two dystrobrevins, five sarcoglycans, five syntrophins, one

dystroglycan and one sarcospan) appear to be reduced to eight in *Drosophila* (one dystrophin, one dystrobrevin, three sarcoglycans, two syntrophins, one dystroglycan and no sarcospan) [46]. Furthermore, *C. elegans* retains all essential human/mouse counterparts of the DGC, but with less diversity [47].

Among the DGC components, only dystrophin and the sarcoglycans are linked to muscular dystrophy. To date, no human mutations have been found in dystroglycan, sarcospan, the syntrophins or the dystrobrevins. Mouse models for each of the core components of the DGC exist, however, and each mouse model (for a summary, see Table 3) will be discussed in relationship to muscular dystrophy.

Animal models for deficiency of dystrophin and dystrophin-binding proteins
Dystrophin

The *mdx* (X-chromosome-linked muscular dystrophy) mouse is the best-characterized mouse model for muscular dystrophy: >500 papers have been published in its analysis.

Table 2

DGC components in various organisms.

Protein	*Mus musculus* Acc No	*Drosophila* Acc No	*C. elegans* Acc No	*Danio rerio* Acc No
Dystroglycan	Dystroglycan X86073	Dystroglycan (DG) AF277390	Dystroglycan Z68011	Dystroglycan BF157619
α-sarcoglycan	α-sarcoglycan AF064081	αε-sarcoglycan (SCGα/ε) AF277391	αε-sarcoglycan AF077544	?
β-sarcoglycan	β-sarcoglycan AF169288	β-sarcoglycan (SGCβ) AF277392	β-sarcoglycan AF099925	β-sarcoglycan AA495110
γ-sarcoglycan	γ-sarcoglycan AF282901	γδ-sarcoglycan (SCGγ/δ) AF277393	γδ-sarcoglycan Z68314	?
δ-sarcoglycan	δ-sarcoglycan AB024923	γδ-sarcoglycan (SCGγ/δ) AF277393	γδ-sarcoglycan Z68314	δ-sarcoglycan AI877617
ε-sarcoglycan	ε-sarcoglycan AF031919	αε-sarcoglycan (SCGα/ε) AF277391	αε-sarcoglycan AF07754	ε-sarcoglycan AW281123
Sarcospan	Sarcospan NM_010656	*Not determined*	?	?
Dystrophin and homologues	Dystrophin, Utrophin, DRP-2 NM_007868 YI2229 U43520	Dystrophin (DYS) X99757	Dystrophin (dys-1) AJ012469	Dystrophin AJ012469
Dystrobrevins	α-, β-dystrobrevin NM_010087 AJ003007	Dystrobrevin (DYB) AF277387	Dystrobrevin (dyb-1) AJ131742	?
Syntrophins	α1-, β1-, β2-syntrophins NM_009228 U89997 U40572[a]	Syntrophin-1 (SYN1) AF277388	β1-syntrophin Z81072	β1-syntrophin AW280633
	γ1-, γ1-syntrophins NM_018967[a] NM_018968[a]	Syntrophin-2 (SYN2) AF277388		

[a] Human sequences. A summary list of DGC sequences from mouse, fruitfly, nematode and zebrafish. Accession (Acc) numbers (underlined) were extracted from GenBank entries for genomic and cDNA clones (see also [46,47]).

As a result of a point mutation in exon 23 of the dystrophin gene, the *mdx* mouse is missing dystrophin [48]. Absence of dystrophin in skeletal muscle also affects the expression of the other DGC components at the sarcolemma [49]. Although this mouse has proved to be a valuable model for DMD, the progressive muscle-wasting disease presents itself in a much milder form than in humans [50] at least in certain muscles. The most affected muscle in the *mdx* mouse is the diaphragm, which reproduces the degenerative changes of muscular dystrophy [51] and the specific twitch force, specific tetanic force and maximum power are all significantly reduced in diaphragm [52]. The *mdx* mice show signs of muscular dystrophy in other muscles during their first six week of life but subsequently show little weakness and have a near-normal lifespan. This partial 'recovery' is as a result of substantial regeneration. The *mdx* mouse adapts to muscle degeneration with an expansion of the satellite cell population and muscle hypertrophy. Transcription factors seem to be important for this successful regeneration. *Mdx* mice lacking the muscle-specific transcription factor MyoD show a more

severe dystrophy due to a deficiency in regenerative capabilities of the muscle [53]. Likewise, *mdx* mice deficient in the myocyte nuclear factor, which is selectively expressed in satellite cells, exhibit an exacerbated dystrophic phenotype as a result of satellite-cell dysfunction [54].

Another possible explanation for the mild phenotype of the *mdx* mouse is that the homologous protein utrophin compensates for the lack of dystrophin. Indeed, mice lacking both dystrophin and utrophin show many signs of DMD in humans: they have a reduced life-span (dying between 4 and 20 weeks), they suffer from severe muscle weakness with joint contractures, pronounced growth retardation, kyphosis and cardiomyopathy, suggesting that dystrophin and utrophin play complementary roles [55,56]. The dystrophic phenotype of the *utrn*^{−/−}/*mdx* mouse was subsequently ameliorated by skeletal-muscle-specific expression of truncated utrophin, indicating that *utrn*^{−/−}/*mdx* mice succumb to a skeletal muscle defect and that their reduced life-span is not as a result of either

Table 3

Summary of DGC-associated mouse models.

Genotype (protein absent)	Life-span	Skeletal dystrophy	Cardiomyopathy
Sgca$^{-/-}$ (α-sarcoglycan)	>1 yr	Moderate	None
Sgcb$^{-/-}$ (β-sarcoglycan)	>1yr	Severe	Severe
Sgcg$^{-/-}$ (γ-sarcoglycan)	20 wks	Severe	Severe
Sgcd$^{-/-}$ (δ-sarcoglycan)	>1yr	Severe	Severe
Sspn$^{-/-}$ (sarcospan)	>1yr	None	None
DG$^{-/-}$ (dystroglycan)	Embryonic lethal	NA	NA
DG chimeric (dystroglycan absent in skeletal and cardiac muscle)	>1yr	Moderate?	Severe
Myd (large)	Reduced	Moderate	None
Mdx (dystrophin)	>1yr	Mild/moderate	Mild
Mdx^{2cv} (dystrophin, Dp260)	>1yr	Mild/moderate	Mild
Mdx^{3cv} (dystrophin, Dp71, Dp116, Dp140, Dp260)	>1yr	Mild/moderate	Mild
Mdx^{4cv} (dystrophin, Dp140, Dp260)	>1yr	Mild/moderate	Mild
Mdx^{5cv} (dystrophin)	>1yr	Mild/moderate	Mild
Mdx52 (dystrophin, Dp140, Dp260)	>1 yr	Mild/moderate	None
Dp71$^{-/-}$	>6 months	None	None
Mnf/mdx (myocyte nuclear factor, Dystrophin)	~3 wks	Severe	NT
MyoD/mdx (MyoD, dystrophin)	1 yr	Severe	Severe
NNOS$^{-/-}$ (neuronal nitric oxide synthase)	>1yr	None	None
nNOS/mdx	>1yr	Mild/moderate	Mild
Utrn$^{-/-}$ (utrophin)	>1yr	None	None
Utrn$^{-/-}$/mdx (utrophin, dystrophin)	4–20 wks	Severe	Severe
Utrn$^{-/-}$/mdx^{3cv} (utrophin, dystrophin, Dp 260, Dp140, Dp116, Dp71)	4–20 wks	Severe	Severe
Adbn$^{-/-}$ (α-dystrobrevin)	>1 yr	Mild	Mild
Adbn$^{-/-}$/mdx (α-dystrobrevin, dystrophin)	8–10 months	Moderate	Moderate
Utrn$^{-/-}$/mdx/adbn$^{-/-}$ (utrophin, dystrophin, α-dystrobrevin)	3–11 wks	Severe	Severe
α1-syntrophin$^{-/-}$	>1yr	None	None
Cav-3$^{-/-}$ (caveolin-3)	>30 wks	Mild	None

NA, not applicable; NT, not tested. (For references, please see main text.)

cardiac or neurogenic components [57]. Although the *mdx* mouse has the capability to adapt to muscle degeneration and utrophin to some extent can compensate for the loss of dystrophin, it should be noted that the serum levels of creatine kinase are elevated in these mice [58], which is a typical diagnostic criteria for muscular dystrophies.

Other than dystrophin, several shorter isoforms are also generated from the dystrophin gene through differential promoter usage. Transcripts from four internal dystrophin promoters produce proteins of 260, 140, 116 and 71 kDa (Dp260, Dp140, Dp116 and Dp71) [59–62]. Dp260 is expressed predominantly in retina [61]. Dp140 is localized in brain and kidney [62,63] and Dp116 is expressed exclusively in Schwann cells [59]. Dp71 is expressed at high levels in almost all tissues except skeletal muscle [60]. Yet, mice with a targeted inactivation of Dp71 display no obvious phenotype [64]. The *mdx*2cv-5cv are other mutant mice generated by chemical mutagenesis using N-ethyl-nitrosurea [65]. The *mdx*3cv has a mutation at the 3′ end of the dystrophin gene, thus resulting in deficiency of the shorter isoforms. Absence of all dystrophin isoforms along with utrophin, however, does not worsen the dystrophic phenotype compared to the *utrn$^{-/-}$/mdx* mouse [66].

Attempts have also been made to produce a dystrophin gene knockout mouse (*mdx*52). Araki *et al*. deleted exon 52 of dystrophin to produce a mouse with a similar phenotype to the *mdx* mouse [67].

Syntrophin

α1-syntrophin is strongly expressed at the sarcolemma and is a core protein of the DGC [7,37]. Yet, mice deficient in α1-syntrophin display no gross histological changes in the skeletal muscle and muscle contractile properties are not altered in these mice [68,69]. nNOS is misplaced from the sarcolemma of these mice and so is aquaporin-4, a mercurial-insensitive water-selective channel [70]. Aquaporin-4 is also absent in skeletal muscle of *mdx* mice [71]. Yet, the role of aquaporin-4 in the pathogenesis of muscular dystrophy is unclear as aquaporin-4-deficient mice maintain normal muscle function [72]. α1-syntrophin is also expressed abundantly at the neuromuscular junction, and recent studies reveal that absence of α1-syntrophin leads to structurally aberrant neuromuscular synapses deficient in utrophin [69].

Dystrobrevin

The best-studied functions of the DGC involve structural stabilization of the sarcolemma: mutations of several DGC components appear to cause muscular dystrophy by disassembling the complex and compromising the linkage between the extracellular matrix of the fibers to its cytoskeleton. Mice deficient in α-dystrobrevin maintain the expression of the DGC at the sarcolemma; yet, these mice develop a mild muscular dystrophy. Analysis of α-dystrobrevin mice revealed that muscular dystrophy might also develop as a result of impaired DGC-dependent signaling. The mechanism behind the disrupted signaling

Figure 2
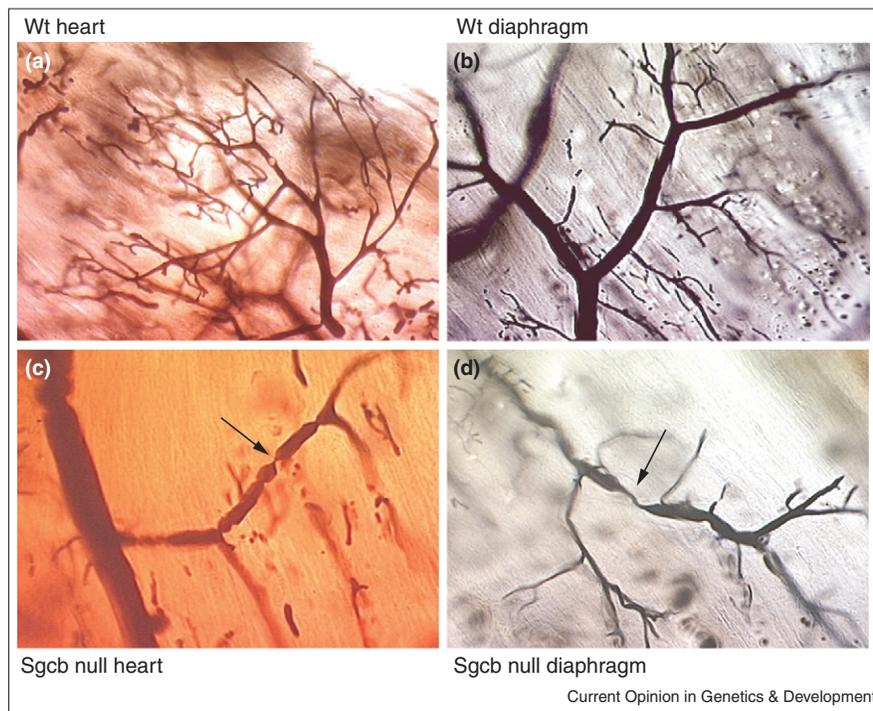
Microfil perfusion of the vasculature.
(a,b) Vessels of 4-week-old heart and diaphragm from wild-type mice show smoothly tapered vessel walls. **(c,d)** In contrast, vessels of the heart and diaphragm from Sgcb-null mice (deficient in β-sarcoglycan) show numerous constrictions (denoted by arrows).

remains to be determined but nNOS may be involved. During exercise, nNOS activity is stimulated to produce cyclic GMP, which is necessary to maintain the blood flow in the active muscle. In electrically stimulated muscle of α-dystrobrevin deficient mice, cyclic GMP levels are not affected in agreement with the finding that nNOS is displaced from the muscle membrane [21]. Yet, loss of nNOS alone is unlikely to account for the dystrophy in α-dystrobrevin-deficient mice, as mice lacking nNOS are not dystrophic [73]. Moreover, the genetic loss of nNOS does not alter the pathogenesis of *mdx* mice [74]. However, recent studies suggest that at least part of the muscle degeneration observed in DMD patients may result from the reduced production of muscle-membrane-associated nNOS. This reduction may lead to improper control of the vasculature and eventual local muscle ischemia [75].

Caveolin-3
Caveolin-3 is a muscle-specific protein integrated in the caveolae, which are small invaginations of the plasma membrane. Mutations in the human caveolin-3 gene cause mild muscular dystrophy (LGMD 1C) [76]. Moreover, caveolin-3 has by immunoprecipitation experiments been shown to interact with dystrophin [77] but it is not an integral component of the DGC [78]. Very recently, caveolin-3-deficient mice were generated [79,80]. These mice exhibit very mild myopathic changes [79,80]. Interestingly, Lisanti and co-workers showed that caveolin-3 expression is required for correct targeting of the DGC to cholesterol-sphingolipid raft domains/caveolae in normal muscle fibers [80].

Animal models for dystroglycan deficiency
Dystroglycan was originally isolated from skeletal muscle but has since been shown to be expressed in a wide variety of tissues and is now considered to be the most broadly expressed DGC component [10,81]. Besides muscle, dystroglycan is expressed at high levels in developing and adult tissues, typically in cell types that adjoin basement membranes such as epithelial and neural tissue [82–84]. Early *in vitro* work demonstrated a role for dystroglycan in epithelial morphogenesis [82]. In 1997, the dystroglycan gene was disrupted in mouse [85]. Dystroglycan null embryos fail to progress beyond the early egg cylinder stage of development and are characterized by structural and functional perturbations of Reichert's membrane, one of the earliest basement membranes that form in the rodent embryo. Subsequent work demonstrated a role for dystroglycan in the formation of the basement membrane of the embryoid body [84]. Because dystroglycan null embryos die early in development prior to gastrulation (i.e. long before any muscle has formed) it is not possible to analyze the consequences of dystroglycan deficiency in muscle. To overcome this, Carbonetto and co-workers generated chimeric mice, lacking dystroglycan in skeletal muscle [86]. Interestingly, these mice develop progressive muscle pathology with changes emblematic to muscular dystrophies in humans. In addition, many neuromuscular junctions are disrupted in these mice. Thus, dystroglycan is necessary for myofiber stability and synapse differentiation or stability. Surprisingly, the basement membrane in the chimeric mice is not grossly perturbed. Yet, the sarcoglycans and dystrophin are absent from the dystroglycan-deficient

muscle fibers suggesting that that the sarcolemmal basement membrane interactions are weakened and/or disorganized.

It is becoming increasingly clear that posttranslational modifications of muscle cell proteins, in particular α-dystroglycan, are important for normal muscle function. Grewal *et al.* [87••] recently presented very intriguing data on the importance of correct glycosylation of α-dystroglycan. The myodystrophy mouse (myd) harbors a mutation in the glycosyltransferase, Large, which leads to altered glyco-sylation of α-dystroglycan (however, with no apparent shift in the molecular weight of α-dystroglycan) and mutant mice develop a progressive myopathy [87••]. Furthermore, muscle–eye–brain disease (MEB) is a type of congenital muscular dystrophy associated with loss-of-function mutations in the gene encoding a glycosyltransferase, POMGnT1 [88••]. POMGnT1 is a glycosylation enzyme that participates in the synthesis of O-mannosyl glycan, a modification that is rare in mammals but is present in α-dystroglycan. Kano *et al.* [89•] recently reported a deficiency of α-dystroglycan, but not β-dystroglycan in MEB patients. A similar selective secondary deficiency of heavily glycosylated α-dystroglycan was also noted in Fukuyama congenital muscular dystrophy (FCMD; in which the gene encoding fukutin is affected) [90]. In summary, a growing body of evidence indicates that some muscular dystrophies may be caused by abnormal glyco-sylation of α-dystroglycan. The mechanistic basis for these disorders is yet to be determined.

Animal models for laminin-2 deficiency

In skeletal muscle, α-dystroglycan binds to the basement membrane protein laminin-2 (composed of laminin α2, β1 and γ1 chains) [24]. About 50% of the patients diagnosed with classical CMD show a primary deficiency of the laminin α2 chain and basement membrane perturbations [91]. Several mouse models for laminin-2 deficiency now exist, including the dy (dystrophia-muscularis) mouse, originally identified at the Jackson Laboratory [24,92,93], and an allelic mutant of the dy mouse, dy^{2J} [94]. Neither of these mouse models exhibits a complete deficiency of laminin α2 chain. Yet, they both display a muscular dystrophy, although the muscular dystrophy in the dy^{2J} presents itself in a milder form compared to the dy mouse. Several laboratories have also generated null mutants for laminin α2 chain (dy^{3K}, dy^W) and all of these mouse models present a severe muscular dystrophy caused by the failure to form a laminin scaffold, which is necessary for basement membrane structure and for interactions with the DGC and integrins [95,96]. Interestingly, a mini agrin gene, which retained high-affinity binding sites for the laminins that are upregulated in dy^W mice (laminin α4 and laminin α5 chains) and α-dystroglycan compensated for the loss of laminin α2 chain [97••]. This suggests that even a non-homologous high-affinity link between dystroglycan and the extracellular matrix is sufficient to prevent muscular dystrophy.

Animal models for integrin deficiency

Laminins also bind integrins, which are a large family of heterodimeric transmembrane cell surface receptors that

Figure 3

Current Opinion in Genetics & Development
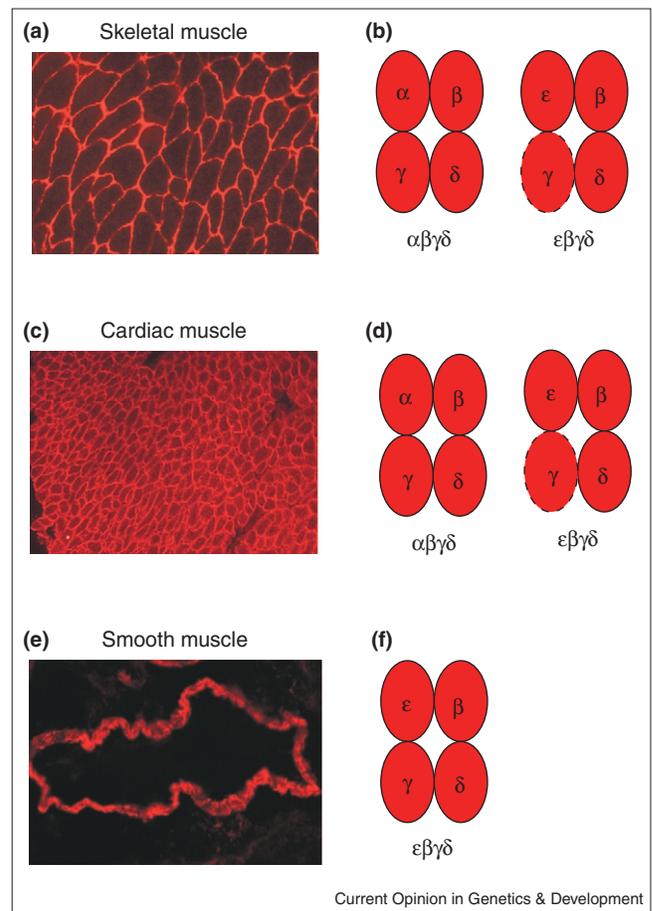
Schematic diagram of sarcoglycan complexes in muscle. **(a)** Mouse skeletal muscle was stained with β-sarcoglycan antibodies. **(b)** Normal skeletal muscle contains two sarcoglycan complexes: an α-sarcoglycan and an ε-sarcoglycan containing complex. The former is composed of α-, β-, γ- and δ-sarcoglycan whereas the latter is composed of at least ε-, β- and δ-sarcoglycan and perhaps γ-sarcoglycan (denoted by a broken line). Note that only the sarcoglycan complexes (and not the entire DGC) are illustrated. **(c)** Mouse cardiac muscle was stained with β-sarcoglycan antibodies. **(d)** Two sarcoglycan complexes are also present within cardiac muscle. **(e)** Pulmonary artery is positively stained with β-sarcoglycan antibodies. **(f)** The smooth muscle sarcoglycan complex is composed of ε-, β-, γ- and δ-sarcoglycan.

function in a wide variety of cell interactions [98]. In skeletal muscle, the α7β1 integrin is the predominant integrin that binds laminin-2 [99]. It is possible that the functions of the DGC and the integrin α7 complex in skeletal muscle to some extent overlap. Mice lacking integrin α7 display a mild myopathy [100] and, interestingly, mice lacking both integrin α7 and dystrophin develop a severe dystrophy and die between 3–4 weeks of age (U Mayer, unpublished data). The fact that the expression of integrin α7 transcript and protein is increased in *mdx* mice and in DMD patients further suggests that the integrin α7β1 may compensate for the absence of the DGC [101,102]. Indeed, transgenic expression of the α7β1 integrin reduces muscular dystrophy and restores viability in the dystrophic *utrn*$^{–/–}$/*mdx* mouse [103].

Although α7β1 integrin is the predominant integrin that binds laminin-2 in skeletal muscle, other integrins are important for normal muscle function too. Mice chimeric for the expression of integrin α5 (receptor for fibronectin) also develop a myopathy [104].

In summary, these data suggest that both the DGC and integrins are involved in anchoring the muscle fiber to its extracellular matrix and that disruption of this anchorage results in muscle instability and subsequently cell death.

Animal models for sarcoglycan–sarcospan deficiency

The sarcoglycan complex is a group of single-pass transmembrane proteins (α-, β-, γ- and δ-sarcoglycan) that is tightly associated with sarcospan to form a subcomplex within the DGC [105]. Although the exact function of the sarcoglycan–sarcospan complex is not known, it is well established that mutations in any of α-, β-, γ- and δ-sarco-glycan genes result in distinct forms of muscular dystrophy now collectively called sarcoglycanopathies [11,14–18,32]. A primary mutation in any one of these genes may lead to either total or partial loss of that sarcoglycan as well as a secondary deficiency of the other sarcoglycans [106]. Furthermore, sarcospan expression is dependent on proper expression of the sarcoglycans [58,105,107]. To date, however, no human mutations have been found in the sarcospan gene, nor have any unclassified muscular dystro-phies been mapped to the chromosomal location of sarcospan [107]. Moreover, sarcospan-deficient mice main-tain their sarcolemmal expression of DGC and maintain normal muscle function [108].

The sarcoglycans are primarily expressed in muscle. α-, β-, γ-, and δ-sarcoglycan are, along with sarcospan, expressed in skeletal and cardiac muscle [109]. In smooth muscle, on the other hand, a unique sarcoglycan–sarcospan complex is expressed. The smooth muscle sarcoglycan complex is composed of ε-sarcoglycan (an α-sarcoglycan homologue) instead of α-sarcoglycan, along with β-sarcoglycan, γ-sarcoglycan and δ-sarcoglycan [81,109,110•]. The sarco-glycan–sarcospan complex is in smooth muscle also associated with dystroglycan. However, dystroglycan in smooth muscle is differentially glycosylated compared to dystroglycan in skeletal muscle [109].

Recent data from several laboratories, including ours, have demonstrated that proper expression of the sarcoglycans in skeletal muscle is a necessity for normal skeletal muscle function [58,111–114,115••]. Furthermore, we have recently demonstrated that proper expression of the sarcoglycans in smooth muscle is also of great importance for normal skeletal and cardiac muscle function [112,115••].

Mice deficient in α-sarcoglycan (Sgca null mice), which is exclusively expressed in striated muscle, develop a progressive muscular dystrophy and a concomitant deficiency in β-, γ- and δ-sarcoglycan along with sarcospan

in skeletal muscle. Furthermore, α-dystroglycan is greatly destabilized at the sarcolemma, indicating that membrane expression of the sarcoglycans is a prerequisite for membrane targeting and stabilization of α-dystroglycan. The sarcoglycan–sarcospan complex is also significantly reduced in cardiac muscle of Sgca null mice. Yet, these mice do not develop cardiomyopathy [58]. The reason for this became clear when β- and δ-sarcoglycan-deficient mice were generated and analyzed in our laboratory [112,115••]. Mice deficient in β- and δ-sarcoglycan (Sgcb null mice and Sgcd null mice, respectively) that are expressed in both striated and smooth muscle also develop a progressive muscular dystrophy, which seems to be more severe than in Sgca null mice. Large, focal areas of necrosis/fibrosis are present in skeletal muscle of Sgcb and Sgcd null mice, a phenomenon that is not detected in Sgca null mice. Both Sgcb and Sgcd null mice also develop cardiomyopathy and morphologically this is recognized by extensive areas of necrosis/fibrosis in the hearts. Additionally, the serum levels for cardiac-specific troponin I are elevated in these mice in agreement with the presence of acute myocardial necrosis that precedes the formation of fibrotic lesions [112,115••,116••]. Furthermore, deficiency of β- and δ-sarcoglycan leads to perturbed expression of the entire sarcoglycan–sarcospan and dystroglycan complexes not only in striated muscle but also in vascular smooth muscle. The loss of the sarco-glycan–sarcospan complex in smooth muscle and the presence of the focal areas of necrosis in skeletal and car-diac muscle prompted us to analyze the blood vessels more closely in Sgcb and Sgcd null mice. Using the Microfil per-fusion technique, we found vascular irregularities in the form of arterial constrictions in both heart and skeletal muscle and also in the kidneys of Sgcb and Sgcd null mice ([112,115••]; M Durbeej, KP Campbell, unpublished data) (see Figure 2). Importantly, the disturbance of the vasculature precedes the onset of ischemic-like lesions. Moreover, no vascular perturbations are present in Sgca null mice. α-sarcoglycan is not expressed in smooth muscle — the smooth muscle sarcoglycan is thus intact in Sgca null mice. Still, Sgca null mice are dystrophic. Together, these data suggest that muscle degeneration does not cause the vascular phenotype. Instead, loss of the sarcoglycan–sar-cospan complex in smooth muscle of blood vessels results in vascular irregularities that aggravate skeletal pathology and initiate heart pathology as a result of diminished delivery of oxygen and nutrients [112,113••,116••].

Biochemical analysis of sarcoglycan deficient mice revealed the presence of a new sarcoglycan complex in skeletal and cardiac muscle ([114,115••]; M Durbeej, KP Campbell, unpublished data). This additional sarcoglycan complex is composed of at least ε-, β-, and δ-sarcoglycan, but neither dystrophin or utrophin [115••] (Figure 3). Thus, β- and δ-sarcoglycan mutations, but not α-sarcoglycan mutations, affect the expression of this complex, which is subsequently absent in Sgcb and Sgcd null mice but not Sgca null mice. Hence, loss of the ε-sarcoglycan complex

may also contribute to the more severe pathology seen in Sgcb and Sgcd null mice. Whether γ-sarcoglycan is part of the ε-sarcoglycan complex is not clear. ε-sarcoglycan expression remains in γ-sarcoglycan-deficient mice [111]. However, immunoprecipitation experiments indicate that γ-sarcoglycan is indeed part of the ε-sarcoglycan complex [114]. Clearly, more studies are needed to clarify this discrepancy.

In summary, a complex pathogenetic mechanism for the development of LGMD 2E and F can be postulated. Deficiency of β- and δ-sarcoglycan causes loss of the sarco-glycan–sarcospan and dystroglycan complexes as well as loss of the ε-sarcoglycan-containing complex in striated muscle. Since the linkage between the extracellular matrix and the muscle cell is perturbed, the skeletal and cardiac muscle membranes become unstable. Thus, the muscle cells may be more prone to ischemic damage that develops as a conse-quence of the absent smooth muscle sarcoglycan complex.

Although α-, β-, γ- and δ-sarcoglycans are part of the same complex, the individual sarcoglycans may exhibit different functional roles. Several lines of evidence support this idea. First, ecto-ATPase activity has been noted for α-sarcoglycan, indicating that α-sarcoglycan might function in controlling the ATP concentration at the surface of the muscle cell [117]. Second, mice deficient for γ-sarcoglycan exhibit a severe muscular dystrophy and cardiomyopathy [111]. Without γ-sarcoglycan, β- and δ-sarcoglycan are unstable at the muscle membrane and α-sarcoglycan is severely reduced. The expression of dystroglycan, however, is not altered and thus the link between the extracellular matrix and the intracellular cytoskeleton appears unaffected. Moreover, γ-sarcoglycan-deficient muscle show normal resistance to mechanical strain, normal peak isometric and tetanic force generation and no evidence for contraction-induced injury after exercise [118]. Thus, a nonmechanical mechanism (perhaps signaling) may be responsible for γ-sarcoglycan-deficient muscular dystrophy. These results are in sharp contrast to muscles of Sgca, Sgcb and Sgcd null mice, which show abnormal resistance to stretch, and a decrease in specific force generation ([58,112]; M Durbeej, KP Campbell, unpublished data).

Conclusions

Animal models lacking each component of the DGC have provided many new insights into the development of muscular dystrophy. Specifically, we have learned that muscular dystrophy can develop when DGC core compo-nents such as sarcoglycans, dystroglycan and dystrobrevins are missing from the skeletal muscle. In addition, muscular dystrophy can also develop when laminin-2 and integrins are absent. More importantly, analysis of these mouse models has unraveled novel pathogenetic mechanisms for the development of muscular dystrophy. For example, perturbation of vascular function together with the disruption of the ε-sarcoglycan complex contribute to the increased severity of mouse models of LGMD type 2E and F.

Furthermore, these animal models will be tremendously valuable tools for testing novel therapeutic approaches. In fact, the feasibility of sarcoglycan gene transfer for sarco-glycanopathies has already been investigated using some of the sarcoglycan-deficient mice. These studies demon-strate that mutations in individual sarcoglycan components can be corrected *in vivo* ([119•,120•]; M Durbeej, KP Campbell, unpublished data). Moreover, the mouse models will be of immense benefit for evaluating possible drug therapies for muscular dystrophy and cardiomyopathy. For example, nicorandil, a vascular smooth muscle relaxant, prevents the acute onset of myocardial necrosis in Sgcd null mice [112]. Furthermore, recent studies in our laboratory suggest that verapamil, a pharmacological agent with vasodilator properties, successfully prevents cardiomyopathy in sarcoglycan-deficient mice by abolishing the vascular dysfunction [116••]. Taken together, the successful treat-ment of cardiomyopathy using agents with clinical relevance in mouse models lacking the smooth muscle sarcoglycan–sarcospan complex connotes that efforts toward drug therapies can be of tremendous benefit preventing certain forms of hereditary cardiomyopathy and perhaps muscular dystrophy.

Acknowledgements

References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:

- • of special interest
- •• of outstanding interest

1. Campbell KP: **Molecular basis of three muscular dystrophies: disruption of the cytoskeleton-extracellular matrix linkage**. *Cell* 1995, **80**:675-679.

2. Straub V, Campbell KP: **Muscular dystrophies and the dystrophin-glycoprotein complex.** *Curr Opin Neurol* 1997, **10**:168-175.

3. Lim LE, Campbell KP: **The sarcoglycan complex in limb-girdle muscular dystrophy.** *Curr Opin Neurol* 1998, **11**:443-452.

4. Bushby K: **The limb-girdle muscular dystrophies-multiple genes, multiple mechanisms.** *Hum Mol Gen* 1999, **8**:1875-1882.

5. Cohn RD, Campbell KP: **The molecular basis of muscular dystrophies**. *Muscle Nerve* 2000, **23**:1456-1471.

6. Hoffman EP, Brown RH Jr, Kunkel: **Dystrophin: the protein product of the Duchenne muscular dystrophy locus**. *Cell* 1987, **51**:919-928.

7. Campbell KP, Kahl SD: **Association of dystrophin and an integral membrane glycoprotein.** *Nature* 1989, **338**:259-262.

8. Ervasti JM, Ohlendieck K, Kahl SD, Campbell KP: **Deficiency of a glycoprotein component of the dystrophin complex in dystrophic muscle.** *Nature* 1990, **345**:315-319.

9. Yoshida M, Ozawa E: **Glycoprotein complex anchoring dystrophin to sarcolemma.** *J Biochem* 1990, **108**:748-752.

10. Ibraghimov-Beskrovnaya O, Ervasti JM, Leveille CJ, Slaughter CA, Sernett SW, Campbell KP: **Primary structure of dystrophin-associated glycoproteins linking dystrophin to the extracellular matrix.** *Nature* 1992, **355**:696-702.

11. Roberds SL, Leturcq F, Allamand V, Piccolo F, Jeanpierre M, Anderson RD, Lim LE, Lee JC, Tome FM, Romero NB *et al.*: **Missense

mutations in the adhalin gene linked to autosomal recessive
muscular dystrophy. *Cell* 1994, **78**:625-633.

12. Adams ME, Butler MH, Dwyer TM, Peters MF, Murnane AA,
    Froehner SC: **Two isoforms of mouse syntrophin, a 58 kd
    dystrophin-associated protein, differ in primary structure and
    tissue distribution.** *Neuron* 1993, **11**:531-540.

13. Yang B, Jung D, Rafael JA, Chamberlain JS, Campbell KP:
    **Identification of alpha-syntrophin binding to the syntrophin triplet,
    dystrophin and utrophin.** *J Biol Chem* 1995, **270**:4975-4978.

14. Lim LE, Duclos F, Broux O, Bourg N, Sunada Y, Allamand V, Meyer J,
    Richard I, Moomaw C, Slaughter C *et al.*: **Beta-sarcoglycan:
    characterization and role in limb-girdle muscular dystrophy linked
    to 4q12.** *Nat Genet* 1995, **11**:257-265.

15. Bönnemann CG, Modi R, Noguchi S, Mizuno Y, Yoshida M,
    Gussoni E, McNally EM, Duggan DJ, Angelini C, Hoffman EP *et al.*:
    **Beta-sarcoglycan (A3b) mutations cause autosomal recessive
    muscular dystrophy with loss of the sarcoglycan complex.** *Nat
    Genet* 1995, **11**:266-273.

16. Noguchi S, McNally EM, Ben Othmane K, Hagiwara Y, Mizuno Y,
    Yoshida M, Yamamoto H, Bonnemann CG, Gussoni E, Denton PH
    et al.: **Mutations in the dystrophin-associated protein
    γ-sarcoglycan in chromosome 13 muscular dystrophy.** *Science*
    1995, **270**:819-822.

17. Nigro V, de Sa Moreira E, Piluso G, Vainzof M, Belsito A, Politano L,
    Puca AA, Passos-Bueno MR, Zatz M: **Autosomal recessive limb-
    girdle muscular dystrophy, LGMD2F, is caused by a mutation in
    the δ-sarcoglycan gene.** *Nat Genet* 1996, **14**:195-198.

18. Jung D, Duclos F, Apostol B, Straub V, Lee JC, Allamand V,
    Venzke DP, Sunada Y, Moomaw CR, Leveille CJ *et al.*:
    **Characterization of δ-sarcoglycan, a novel component of the
    oligomeric sarcoglycan complex involved in limb-girdle muscular
    dystrophy.** *J Biol Chem* 1996, **271**:32321-32329.

19. Crosbie RH, Heighway J, Venzke DP, Lee JC, Campbell KP:
    **Sarcospan, the 25 kDa transmembrane component of the
    dystrophin-glycoprotein complex.** *J Biol Chem* 1997,
    272:31221-31224.

20. Piluso G. Mirabella M, Ricci E, Belsito A, Abbondanza C, Servidei S,
    Puca AA, Tonali P, Puca GA, Nigro V: **γ1- and γ2-syntrophins, two
    novel dystrophin binding proteins localized in neuronal cells.**
    J Biol Chem 2000, **275**:15851-15860.

21. Grady RM, Grange RW, Lau KS, Maimone MM, Nichol MC, Stull JT,
    Sanes JR: **Role for the α-dystrobrevin in the pathogenesis of
    dystrophin dependent muscular dystrophies.** *Nat Cell Biol* 1999,
    4:215-220.

22. Ervasti JM, Campbell KP: **Membrane organization of the
    dystrophin-glycoprotein complex.** *Cell* 1991, **66**:1121-1131.

23. Ervasti JM, Campbell KP: **A role for dystrophin-glycoprotein
    complex as a transmembrane linker between laminin and actin.**
    J Cell Biol 1993, **122**:809-823.

24. Sunada Y, Bernier SM, Kozak CA, Yamada Y, Campbell KP:
    **Deficiency of merosin in dystrophic dy mice and genetic linkage
    of laminin M chain to dy locus.** *J Biol Chem* 1994,
    269:13729-13732.

25. Campanelli JT, Roberds SL, Campbell KP, Scheller R: **A role for
    dystrophin associated glycoproteins and utrophin in agrin-
    induced AChR clustering.** *Cell* 1994, **77**:663-674.

26. Gee SH, Montanaro F, Lindenbaum MH, Carbonetto S:
    **Dystroglycan-α, a dystrophin-associated glycoprotein, is a
    functional agrin receptor.** *Cell* 1994, **77**:675-686.

27. Talts JF, Andac Z, Gohring W, Brancaccio A, Timpl R: **Binding of the
    G domains of laminin α1 and α2 chains and perlecan to heparin,
    sulfatides, α-dystroglycan and several extracellular matrix
    proteins.** *EMBO J* 1999, **18**:863-870.

28. Bowe MA, Mendis DB, Fallon JR: **The small leucine-rich repeat
    proteoglycan biglycan binds to α-dystroglycan and is upregulated
    in dystrophic muscle.** *J Cell Biol* 2000, **146**:801-810.

29. Sugita S, Saito F, Tang, J, Satz J, Campbell, KP, Sudhof TC:
    A stochiometric complex of neurexins and dystroglycan in brain.
    J Cell Biol 2001, **154**:435-445.

30. Weller B, Karpati G, Carpenter S: **Dystrophin-deficient mdx muscle
    fibers are preferentially vulnerable to necrosis induced by**

31. Petrof BJ, Shrager JB, Stedman HH, Kelly AM, Sweeney HL:
    **Dystrophin protects the sarcolemma from stresses developed
    during muscle contraction.** *Proc Natl Acad Sci USA* 1993,
    90:3710-3714.

32. Piccolo F, Roberds SL, Jeanpierre M, Leturcq F, Azibi K, Beldjord C,
    Carrie A, Recan D, Chaouch M, Reghis A *et al.*: **Primary
    adhalinopathy: a common cause of autosomal recessive
    muscular dystrophy of variable severity.** *Nat Genet* 1995,
    10:243-245.

33. Allamand V, Sunada Y, Salih MA, Straub V, Ozo CO, Al-Turaiki MH,
    Akbar M, Kolo T, Colognato H, Zhang X, Sorokin LM *et al.*: **Mild
    congenital muscular dystrophy in two patients with an internally
    deleted laminin alpha2-chain.** *Hum Mol Genet* 1997, **6**:747-752.

34. Yang B, Jung D, Motto D, Meyer J, Koretzky G, Campbell KP: **SH3
    domain mediated interaction of dystroglycan and Grb2.** *J Biol
    Chem* 1995, **270**:11711-11714.

35. Thompson TG, Chan YM, Hack AA, Brosius M, Rajala M, Lidov HG,
    McNally EM, Watkins S, Kunkel LM: **Filamin 2 (FLN2): a muscle-
    specific sarcoglycan interacting protein.** *J Cell Biol* 2000,
    148:115-126.

36. Ahn AH, Freener CA, Gussoni E, Yoshida M, Ozawa E, Kunkel LM:
    **The three human syntrophin genes are expressed in diverse
    tissues, have distinct chromosomal locations, and each bind to
    dystrophin and its relatives.** *J Biol Chem* 1996, **271**:2724-2730.

37. Peters, MF, Adams ME, Froehner SC: **Differential association of
    syntrophin pairs with the dystrophin complex.** *J Cell Biol* 1997,
    138:81-93.

38. Brenman JE, Chao DS, Gee SH, McGee AW, Craven SE,
    Santillano DR, Wu Z, Huang F, Xia H, Peters MF, Froehner SC *et al.*:
    **Interaction with nitric oxide synthase with the postsynaptic
    density protein PSD-95 and α1-syntrophin mediated by PZD
    domains.** *Cell* 1996, **84**:757-767.

39. Schultz J, Hoffmuller U, Krause G, Ashurst J, Macias MJ, Schmieder P,
    Schneider-Mergener J, Oschkinat H: **Specific interactions between
    the syntrophin PDZ domain and the voltage-gated sodium
    channels.** *Nature Struct Biol* 1998, **5**:19-24.

40. Gee SH, Madhavan R, Levinson SR, Caldwell JH, Sealock R,
    Froehner SC: **Interaction of brain and muscle sodium channels
    with multiple members of the syntrophin family of dystrophin-
    associated proteins.** *J Neurosci* 1998, **18**:128-137.

41. Lumeng C, Phelps S, Crawford GE, Walden PD, Barald K,
    Chamberlain JS: **Interactions between β2-syntrophin and family of
    microtubule-associated serine/threonine kinases.** *Nat Neurosci*
    1999, **2**:611-617.

42. Wagner KR, Cohen JB, Huganir RL: **The 87K postsynaptic
    membrane protein from Torpedo is a protein-tyrosine kinase
    substrate homologous to dystrophin.** *Neuron* 1993, **10**:511-522.

43. Sadoulet-Puccio HM, Rajala M, Kunkel LM: **Dystrobrevin and
    dystrophin: an interaction through coiled-coil motifs.** *Proc Natl
    Acad Sci USA* 1997, **94**:12413-12418.

44. Yoshida M, Hama H, Ishikawa-Sakurai M, Imamura M, Mizuno Y,
    Araishi K, Wakabayashi-Takai E, Noguchi S, Sasaoka T, Ozawa E:
    **Biochemical evidence for association of dystrobrevin with the
    sarcoglycan-sarcospan complex as a basis for understanding
    sarcoglycanopathies.** *Hum Mol Genet* 2000, **9**:1033-1040.

45. Roberts RG, Bobrow M: **Dystrophins in vertebrates and
    invertebrates.** *Hum Mol Gen* 1998, **79**:589-595.

46. Greener MJ, Roberts RG: **Conservation of components of the
    dystrophin complex in *Drosophila*.** *FEBS Lett* 2000, **482**:13-18.

47. Hutter H, Vogel BE, Plenefisch JD, Norris CR, Proenca RB, Spieth J,
    Guo C, Mastwal S, Zhu X, Scheel J *et al.*: **Conservation and novelty
    in the evolution of cell adhesion and extracellular matrix genes.**
    Science 2000, **287**:989-994.

48. Sicinski P, Geng Y, Ryder-Cook AS, Barnard EA, Darlison MG,
    Barnard PJ: **The molecular basis of muscular dystrophy in the mdx
    mouse: a point mutation.** *Science* 1989, **244**:1578-1580.

49. Ohlendieck K, Campbell, KP: **Dystrophin-associated proteins are
    greatly reduced in skeletal muscle from mdx mice.** *J Cell Biol*
    1991, **115**:1685-1694.

experimental lengthening contractions. *J Neurol Sci* 1990,
100:9-13.

50.   Bulfield G, Siller WG, Wight PA, Moore KJ: **Skeletal muscle pathology in X chromosome-linked muscular dystrophy (mdx) in the mouse**. *Proc Natl Acad Sci USA* 1984, **81**:1189-1192.

51.   Stedman H Sweeney HL, Shrager JB, Maguire HC, Panettieri RA, Petrof B, Narusawa M, Leferovich JM, Sladky JT, Kelly AM: **The mdx diaphragm reproduces the degenerative changes of Duchenne muscular dystrophy**. *Nature* 1991, **352**:536-539.

52.   Stevens ED, Faulkner JA: **The capacity of mdx mouse diaphragm muscle to do oscillatory work**. *J Physiol* 2000, **1**:457-466.

53.   Megeney LA, Kablar B, Garrett, K, Anderson JE, Rudnicki MA: **MyoD is required for myogenic stem cell function in adult skeletal muscle**. *Genes Dev* 1996, **10**:1173-1183.

54.   Garry DJ, Meeson A, Elterman J, Zhao Y, Yang P, Bassel-Duby R, Williams RS: **Myogenic stem cell function is impaired in mice lacking the forkhead/winged helix protein MNF**. *Proc Natl Acad Sci USA* 2000, **97**:5416-5421.

55.   Deckoninck AE, Rafael JA, Skinner JA, Brown SC, Potter AC, Metzinger L, Watt DJ, Dickson JG, Tinsley JM, Davies KE: **Utrophin-dystrophin-deficient mice as a model for Duchenne Muscular Dystrophy**. *Cell* 1997, **90**:7171-727.

56.   Grady RM, Teng H, Nichol, M, Cunningham C, Wilkinson RS, Sanes JR: **Skeletal and cardiac myopathies in mice lacking utrophin and dystrophin: a model for Duchenne muscular dystrophy**. *Cell* 1997, **90**:729-738.

57.   Rafael JA, Tinsley JM, Potter AC, Deconinck AE, Davies KE: **Skeletal muscle-specific expression of a utrophin transgene rescues utrophin-dystrophin deficient mice**. *Nat Genet* 1998, **19**:79-82.

58.   Duclos F, Straub V, Moore SA, Venzke DP, Hrstka RF, Crosbie RH, Durbeej M, Lebakken CS, Ettinger AJ, van der Meulen J *et al.*: **Progressive muscular dystrophy in α-sarcoglycan deficient mice**. *J Cell Biol* 1998, **142**:1461-1471.

59.   Byers TJ, Lidov HG, Kunkel LM: **An alternative dystrophin transcript specific to peripheral nerve**. *Nat Genet* 1993, **4**:87-93.

60.   Schofield J, Blake DJ, Simmons C, Morris GE, Tinsley JM, Davies KE, Edwards YH: **Apo-dystrophin-1 and apo-dystrophin-2, products of the Duchenne muscular dystrophy locus: expression during mouse embryogenesis and in cultured cell lines**. *Hum Mol Genet* 1994, **8**:1309-1316.

61.   D'Souza V, Nguyen TM, Morris GE, Karges W, Pillers DA, Ray PN: **A novel dystrophin isoforms is required for normal retinal electrophysiology**. *Hum Mol Genet* 1995, **4**:837-842.

62.   Lidov HG, Selig S, Kunkel LM: **Dp140: a novel 140 kDa CNS transcript from the dystrophin locus**. *Hum Mol Genet* 1995, **3**:329-335.

63.   Durbeej M, Jung D, Hjalt T, Campbell KP, Ekblom P: **Transient expression of Dp140, a product of the Duchenne muscular dystrophy locus, during kidney tubulogenesis**. *Dev Biol* 1997, **181**:156-167.

64.   Sarig R, Sarig R, Mezger-Lallemand V, Gitelman I, Davis C, Fuchs O, Yaffe D, Nudel U: **Targeted inactivation of Dp71, the major non-muscle product of the DMD gene: differential activity of the Dp71 promoter during development**. *Hum Mol Genet* 1999, **8**:1-10.

65.   Im WB, Phelps SF, Copen EH, Adams EG, Slightom JL, Chamberlain JS: **Differential expression of dystrophin isoforms in strains of mdx mice with different mutations**. *Hum Mol Genet* 1996, **8**:1149-1153.

66.   Rafael JA, Trickett JI, Potter AC, Davies KE: **Dystrophin and utrophin do not play crucial roles in nonmuscle tissues in mice**. *Muscle Nerve* 1999, **22**:517-519.

67.   Araki E, Nakamura K, Nakao K, Kameya S, Kobayashi O, Nonaka I, Kobayashi T, Katsuki M: **Targeted disruption of exon 52 in the mouse dystrophin gene induced muscle degeneration similar to that observed in Duchenne muscular dystrophy**. *Biochem Biophys Res Com* 1997, **238**:492-497.

68.   Kameya S, Miyagoe Y, Nonaka I, Ikemoto T, Endo M, Hanaoka K, Nabeshima Y, Takeda S: **α1-syntrophin gene disruption results in the absence of neuronal-type nitric-oxide synthase at the sarcolemma but does not induce muscle degeneration**. *J Biol Chem* 1999, **274**:2193-2200.

69.   Adams M, ME, Kramarcy N, Krall SP, Rossi SG, Rotundo RL, Sealock R, Froehner SC: **Absence of α-syntrophin leads to**

70.   Yokota T, Miyagoe Y, Hosaka Y, Tsukita K, Kameya S, Shibuya S, Matsuda R, Wakayama Y, Takeda S: **Aquaporin-4 is absent at the sarcolemma and at perivascular astrocyte endfeet in α1-syntrophin knockout mice**. *Proc Japan Acad* 2000, **76**:22-27.

71.   Liu JW, Wakayama Y, Inoue M, Shibuya S, Kojima H, Jimi T, Oniki H: **Immunocytochemical studies of aquaporin-4 in the skeletal muscle**. *J Neurol Sci* 1999, **15**:24-28.

72.   Yang B, Verbavatz JM, Song Y, Vetrivel L, Manley G, Kao WM, Ma T, Verkman AS: **Skeletal muscle function and water permeability in aquaporin-4 deficient mice**. *Am J Physiol Cell Physiol* 2000, **278**:1109-1105.

73.   Huang PL, Dawson TM, Bredt DS, Snyder SH, Fishman MC: **Targeted disruption of the nitric oxide synthase gene**. *Cell* 1993, **75**:1273-1280.

74.   Crosbie RH Straub V, Yun HY, Lee JC, Rafael JA, Chamberlain JS, Dawson VL, Dawson TM, Campbell KP: **mdx muscle pathology is independent of nNOS perturbation**. *Hum Mol Genet* 1998, **7**:823-829.

75.   Sander M, Chavoshan B, Harris SA, Iannaccone ST, Stull JT, Thomas GD, Victor RG: **Functional muscle ischemia in neuronal nitric oxide synthase deficient skeletal muscle of children with Duchenne muscular dystrophy**. *Proc Natl Acad Sci USA* 2000, **5**:13818-13823.

76.   Minetti C, Sotgia F, Bruno C, Scartezzini P, Broda P, Bado M, Masetti E, Mazzocco M, Egeo A, Donati MA *et al.*: **Mutations in the caveolin-3 gene cause autosomal dominant limb-girdle muscular dystrophy**. *Nat Genet* 1998, **18**:365-368.

77.   Song KS, Scherer PE, Tang Z, Okamoto T, Li S, Chafel M, Chu C, Kohtz DS, Lisanti MP: **Expression of caveolin-3 in skeletal, cardiac, and smooth muscle cells: caveolin-3 is a component of the sarcolemma and co-fractionates with dystrophin and dystrophin-associated proteins**. *J Biol Chem* 1996, **271**:15160-15165.

78.   Crosbie RH, Yamada H, Venzke DP, Lisanti MP, Campbell KP: **Caveolin-3 is not an integral component of the dystrophin–glycoprotein complex**. *FEBS Lett* 1998, **427**:279-282.

79.   Hagiwara Y, Sasaoka T, Araishi K, Imamura M, Yorifuji H, Nonaka I, Ozawa E, Kikuchi T: **Caveolin-3 deficiency causes muscle degeneration in mice**. *Hum Mol Genet* 2000, **9**:3047-3054.

80.   Galbiati F, Engelman JA, Volonte D, Zhang XL, Minetti C, Li M, Hou H Jr, Kneitz B, Edelmann W, Lisanti MP: **Caveolin-3 null mice show a loss of caveolae, changes in the microdomain distribution of the dystrophin–glycoprotein complex, and T-tubule abnormalities**. *J Biol Chem* 2001, **276**:21425-21433.

81.   Durbeej M, Campbell KP: **Biochemical characterization of the epithelial dystroglycan complex**. *J Biol Chem* 1999, **274**:26609-26616.

82.   Durbeej M, Larsson E, Ibraghimov-Beskrovnaya O, Roberds SL, Campbell KP, Ekblom P: **Non-muscle α-dystroglycan is involved in epithelial development**. *J Cell Biol* 1995, **130**:79-91.

83.   Durbeej M, Henry MD, Ferletta M, Campbell KP, Ekblom P: **Distribution of dystroglycan in normal adult mouse tissues**. *J Histochem Cytochem* 1998, **46**:449-457.

84.   Henry MD, Campbell KP: **A role for dystroglycan in basement membrane assembly**. *Cell* 1998, **95**:859-870.

85.   Williamson RA, Henry MD, Daniels KJ, Hrstka RF, Lee JC, Sunada Y, Ibraghimov-Beskrovnaya O, Campbell K: **Dystroglycan is essential for early embryonic development: disruption of Reichert's membrane in Dag1-null mice**. *Hum Mol Genet* 1997, **6**:831-841.

86.   Côté PD, Moukhles H, Lindenbaum M, Carbonetto S: **Chimeric mice deficient in dystroglycans develop muscular dystrophy and have disrupted monaural synapses**. *Nat Genet* 1999, **23**:338-342.

87.   Grewal P, Holzfeind PJ, Bittner RE, Hewitt JE: **Mutant**
••   **glycosyltransferase and altered glycosylation of α-dystroglycan in the myodystrophy mouse**. *Nat Genet* 2001, **28**:151-154.
The authors show that the muscle-wasting phenotype in this animal model is caused by loss-of-function mutations in LARGE, encoding a glycosyltrans-ferase. Importantly, glycosylation of α-dystroglycan is altered in the myd mouse, suggesting that abnormal glycosylation of α-dystroglycan may be the basis for a novel 'pathomechanism' for some muscular dystrophies.

88. Yoshida A, Kobayashi K, Manya H, Taniguchi K, Kano H, Mizuno M,
•• Inazu T, Mitsuhashi H, Takahashi S, Takeuchi M *et al.*: **Muscular dystrophy and neuronal migration disorder caused by mutations in a glycosyltransferase, POMGnT1.** *Dev Cell* 2001, **1**:717-724.
An important paper elucidating the genetic basis for MEB. Loss-of-function mutations are described in the glycosyltransferase POMGnT1, an enzyme that participates in O-mannosyl glycan synthesis. Mammalian O-mannosyl glycosylation is a rare type of post-translational modification but interestingly it occurs on α-dystroglycan. Together with [87••], these data indicate that altered glycosylation of α-dystroglycan is one cause of muscular dystrophies. See also [89•].

89. Kano H, Kobayashi K, Herrmann R, Tachikawa M, Manya H, Nishino I,
• Nonaka I, Straub V, Talim B, Voit T *et al.*: **Deficiency of α-dystroglycan in muscle-eye-brain disease.** *Biochem Biophys Res Comm* 2002, **291**:1283-1286.
A follow up study to [88••]. Immunohistochemical and immunoblot analyses demonstrate that α-dystroglycan, but not β-dystroglycan is absent in skeletal muscle of MEB patients.

90. Hayashi YK, Ogawa M, Tagawa K, Noguchi S, Ishihara T, Nonaka I, Arahata K: **Selective deficiency of alpha-dystroglycan in Fukuyama-type congenital muscular dystrophy.** *Neurology* 2001, **10**:115-121.

91. Tomé FM: **The Peter Emil Becker Award lecture: The saga of congenital muscular dystrophy.** *Neuropediatrics* 1998, **30**:55-65.

92. Michelson AM, Russell E, Harman PJ: **Dystrophia muscularis: a hereditary primary myopathy in the house mouse.** *Proc Acad Natl Sci USA* 1955, **41**:1079-1084.

93. Xu H, Christmas P, Wu X, Wewer U, Engvall, E: **Defective muscle basement membrane and lack of M-laminin in the dy/dy mouse.** *Proc Natl Acad Sci USA* 1994, **91**:5572-5576.

94. Xu H, Wu R, Wewer U, Engvall E: **Murine muscular dystrophy caused by a mutation in the laminin alpha 2 (Lama2) gene.** *Nat Genet* 1994, **8**:297-302.

95. Miyagoe Y, Hanaoka K, Nonaka I, Hayasaka M, Nabeshima Y, Arahata K, Nabeshima Y, Takeda S: **Laminin alpha2 chain-null mutant mice by targeted disruption of the Lam2 gene: a new model of merosin (laminin 2)-deficient congenital muscular dystrophy.** *FEBS Lett* 1997, **415**:33-39.

96. Kuang W, Xu H, Vachon PH, Liu L, Loechel F, Wewer UM, Engvall E: **Merosin-deficient congenital muscular dystrophy. Partial genetic correction in two mouse models.** *J Clin Invest* 1998, **102**:844-852.

97. Moll J, Barzaghi P, Lin S, Bezakova G, Lochmuller H, Engvall E,
•• Muller U, Ruegg MA: **An agrin minigene rescues dystrophic symptoms in a mouse model for congenital muscular dystrophy.** *Nature* 2001, **413**:302-307.
An intriguing approach to alleviate muscle pathology in the laminin-α2-deficient dyw mouse. An agrin minigene was designed to restore the link between dystroglycan and the extracellular matrix. Transgenic overexpression of the minigene in the skeletal muscles of dyw mice prevented muscle wasting. These data provide evidence that a non-homologous protein in combination with a rational protein design could replace the function of a missing protein.

98. Hynes RO: **Integrins: versatility, modulation, and signaling in cell adhesion.** *Cell* 1992, **69**:11-25.

99. Burkin DJ, Kaufman SJ: **The α7β1 integrin in muscle development and disease.** *Cell Tiss Res* 1999, **296**:183-190.

100. Mayer U, Saher G, Fassler R, Bornemann A, Echtermeyer F, von der Mark H, Miosge N, Poschl E, von der Mark K: **Absence of integrin α7 causes a novel form of muscular dystrophy.** *Nat Genet* 1997, **17**:318-323.

101. Hodges BL, Hayashi YK, Nonaka I, Wang W, Arahata K, Kaufman SJ: **Altered expression of the α7β1 integrin in human and murine muscular dystrophies.** *J Cell Sci* 1997, **110**:2873-2881.

102. Cohn RD, Mayer U, Saher G, Herrmann R, van der Flier A, Sonnenberg A, Sorokin L, Voit T: **Secondary reduction of alpha7B integrin in laminin α2 deficient congenital muscular dystrophy supports an additional transmembrane link in skeletal muscle.** *J Neurol Sci* 1999, **163**:140-152.

103. Burkin DJ, Wallace GQ, Nicol KJ, Kaufman DJ, Kaufman SJ: **Enhanced expression of the alpha 7 beta 1 integrin reduces muscular dystrophy and restores viability in dystrophic mice.** *J Cell Biol* 2001, **152**:1207-1218.

104. Taverna D, Disatnik MH, Rayburn H, Bronson RT, Yang J, Rando TA, Hynes RO: **Dystrophic muscle in mice chimeric for expression of α5 integrin.** *J Cell Biol* 1998, **143**:849-859.

105. Crosbie RH, Lebakken CS, Holt KH, Venzke DP, Straub V, Lee JC, Grady RM, Chamberlain JS, Sanes JR, Campbell KP: **Membrane targeting and stabilization of sarcospan is mediated by the sarcoglycan subcomplex.** *J Cell Biol* 1999, **145**:153-165.

106. Barresi R, Confalonieri V, Lanfossi M, Di Blasi C, Torchiana E, Mantegazza R, Jarre L, Nardocci N, Boffi P, Tezzon F *et al.*: **Concomitant deficiency of β- and γ-sarcoglycans in 20 α-sarcoglycan (adhalin)-deficient patients: immunohistochemical analysis and clinical aspects.** *Acta Neuropathol* 1997, **94**:28-35.

107. Crosbie RH, Im LE, Moore SA, Hirano M, Hays AP, Maybaum SW, Collin H, Dovico SA, Stolle CA, Fardeau M *et al.*: **Molecular and genetic characterization of sarcospan: insights into sarcoglycan–sarcospan domains.** *Hum Mol Genet* 2000, **13**:2019-2023.

108. Lebakken CS, Venzke DP, Hrstka RF, Consolino CM, Faulkner JA, Williamson RA, Campbell KP: **Sarcospan-deficient mice maintain normal muscle function.** *Mol Cell Biol* 2000, **20**:1669-1677.

109. Straub V, Ettinger AJ, Durbeej M, Venzke DP, Cutshall S, Sanes JR, Campbell KP: **ε-sarcoglycan replaces α-sarcoglycan in smooth muscle to form a unique dystrophin-glycoprotein complex.** *J Biol Chem* 1999, **274**:27989-27996.

110. Barresi R, Moore SA, Stolle CA, Mendell JR, Campbell KP:
• **Expression of γ-sarcoglycan in smooth muscle and its interaction with the smooth muscle sarcoglycan-sarcospan complex.** *J Biol Chem* 2000, **275**:38554-38560.
The expression pattern of γ-sarcoglycan in smooth muscle is biochemically and genetically evaluated in this paper as it had been unclear whether γ-sarcoglycan is expressed in smooth muscle. The authors provide convincing evidence that γ-sarcoglycan is a component of the smooth muscle sarcoglycan complex.

111. Hack AA, Ly CT, Jiang F, Clendenin CJ, Sigrist KS, Wollmann RL, McNally EM: **γ-sarcoglycan deficiency leads to muscle membrane defects and apoptosis independent of dystrophin.** *J Cell Biol* 1998, **142**:1279-1287.

112. Coral-Vasquez R, Cohn RD, Moore SA, Hill JA, Weiss RM, Davisson RL, Straub V, Barresi R, Bansal D, Hrstka RF *et al.*: **Disruption of the sarcoglycan-sarcospan complex in vascular smooth muscle: a novel mechanism for cardiomyopathy and muscular dystrophy.** *Cell* 1999, **98**:465-474.

113. Araishi K, Sasaoka T, Imamura M, Noguchi S, Hama H, Wakabayashi E, Yoshida M, Hori T, Ozawa E: **Loss of the sarcoglycan complex and sarcospan leads to muscular dystrophy in beta-sarcoglycan deficient mice.** *Hum Mol Genet* 1999, **8**:1589-1598.

114. Liu AL, Engvall E: **Sarcoglycan isoforms in skeletal muscle.** *J Biol Chem* 1999, **272**:38171-38176.

115. Durbeej M, Cohn RD, Hrstka RF, Moore SA, Allamand V,
•• Davidson BL, Williamson RA, Campbell KP: **Disruption of the β-sarcoglycan gene reveals pathogenetic complexity of limb-girdle muscular dystrophy type 2E.** *Mol Cell* 2000, **5**:141-151.
Together with [112], the findings suggest a novel pathogenetic mechanism whereby disruption of the sarcoglycan–sarcospan complex in vascular smooth muscle causes disturbance of the vasculature, which in turn leads to the development of a severe cardiomyopathy and exacerbation of muscular dystrophy in β- and δ-sarcoglycan deficient mice. The presence of a distinct ε-sarcoglycan complex in skeletal muscle is also presented.

116. Cohn R, Durbeej M, Moore SA, Coral-Vazquez R, Prouty S,
•• Campbell KP: **Prevention of cardiomyopathy in mouse models lacking the smooth muscle sarcoglycan-sarcospan complex.** *J Clin Invest* 2001, **107**:R1-R9.
An interesting study showing that pharmacological intervention using oral treatment with verapamil is able to prevent cardiomyopathy in mice with disruption of the smooth muscle sarcoglycan complex.

117. Betto R, Senter L, Ceoldo S, Tarricone E, Biral D, Salviati G: **Ecto-ATPase activity of α-sarcoglycan (adhalin).** *J Biol Chem* 1999, **274**:7907-7912.

118. Hack AA, Cordier L, Shoturma DI, Lam MY, Sweeney HL, McNally EM: **Muscle degeneration without mechanical injury in sarcoglycan deficiency.** *Proc Natl Acad Sci USA* 1999, **96**:10723-10728.

119. Allamand V, Donahue KM, Straub V, Davisson RL, Davidson BL,
• Campbell KP: **Early adenovirus-mediated gene transfer effectively prevents muscular dystrophy in alpha-sarcoglycan deficient mice.** *Gene Therapy* 2000, **7**:1385-1391.
This paper shows that early adenovirus-mediated gene transfer effectively prevents muscular dystrophy in α-sarcoglycan deficient mice. One single injection of a first generation adenovirus into the skeletal muscle of neonate mice led to sustained expression of α-sarcoglycan at the sarcolemma of transduced fibers for at least 7 months.

120. Cordier L, Hack AA, Scott MO, Barton-Davis ER, Gao G, Wilson JM,
• McNally EM, Sweeney HL: **Rescue of skeletal muscles of γ-sarcoglycan-deficient mice with adeno-associated virus-mediated gene transfer.** *Mol Therapy* 2000, **1**:119-129.
The authors demonstrate that injections of a recombinant adeno-associated virus – in which human γ-sarcoglycan is driven by a muscle creatine kinase promoter – into skeletal muscle of γ-sarcoglycan-deficient mice result in an overall improvement of the histological pattern of dystrophy. However, the authors underscore the need for intervention early in the time course of the disease process.

121. Sullivan T, Escalante-Alcalde D, Bhatt H, Anver M, Bhat N, Nagashima K, Stewart CL, Burke B: **Loss of A-type lamin expression compromises nuclear envelope integrity leading to muscular dystrophy.** *J Cell Biol* 1999, **147**:913-920.

122. Richard I, Roudaut C, Marchand S, Baghdiguian S, Herasse M, Stockholm D, Ono Y, Suel L, Bourg N, Sorimachi H *et al.*: **Loss of calpain 3 proteolytic activity leads to muscular dystrophy and to apoptosis-associated IκBα/nuclear factor κB pathway perturbation in mice.** *J Cell Biol* 2000, **151**:1583-1590.

123. Bittner RE, Anderson LV, Burkhardt E, Bashir R, Vafiadaki E, Ivanova S, Raffelsberger T, Maerk I, Hoger H, Jung M *et al.*: **Dysferlin deletion in SJL mice (SJL-Dysf) defines a natural model for limb girdle muscular dystrophy 2B.** *Nat Genet* 1999, **23**:141-142.

124. Frosk P, Weiler T, Nylen E, Sudha T, Greenberg CR, Morgan K, Fujiwara TM, Wrogemann K: **Limb-girdle muscular dystrophy type 2H associated with mutation in TRIM32, a putative E3-ubiquitin-ligase gene.** *Am J Hum Genet* 2002, **70**:663-672.

125. Brockington M, Yuva Y, Prandini P, Brown SC, Torelli S, Benson MA, Herrmann R, Anderson LV, Bashir R, Burgunder JM *et al.*: **Mutations in the fukutin-related protein identify limb-girdle muscular dystrophy 2I as a milder allelic variant of congenital muscular dystrophy MDC1C.** *Hum Mol Genet* 2001, **10**:2851-2859.

126. Camacho Vanegas O, Bertini E, Zhang RZ, Petrini S, Minosse C, Sabatelli P, Giusti B, Chu ML, Pepe G: **Ullrich scleroatonic muscular dystrophy is caused by recessive mutations in collagen typeVI.** *Proc Natl Acad Sci USA* 2001, **98**:7516-7521.

127. Cormand B, Pihko H, Bayes M, Valanne L, Santavuori P, Talim B, Gershoni-Baruch R, Ahmad A, van Bokhoven H, Brunner HG *et al.*: **Clinical and genetic distinction between Walker-Warburg syndrome and muscle-eye-brain disease.** *Neurology* 2001, **56**:1059-1069.

128. Moghadaszadeh B, Petit N, Jaillard C, Brockington M, Roy SQ, Merlini L, Romero N, Estournet B, Desguerre I, Chaigne D *et al.*: **Mutations in SEPN1 cause congenital muscular dystrophy with spinal rigidity and restrictive respiratory syndrome.** *Nat Genet* 2001, **29**:17-18.

129. Bonaldo P, Braghetta P, Zanetti M, Piccolo S, Volpin D, Bressan GM: **Collagen VI deficiency induces early onset myopathy in the mouse: an animal model for Bethlem myopathy.** *Hum Mol Genet* 1998, **7**:2135-2140.

130. Eklund L, Piuhola J, Komulainen J, Sormunen R, Ongvarrasopone C, Fassler R, Muona A, Ilves M, Ruskoaho H, Takala TE *et al.*: **Lack of type XV collagen causes a skeletal myopathy and cardiovascular defects in mice.** *Proc Natl Acad Sci USA* 2001, **98**:1194-1199.

131. Andra K, Lassmann H, Bittner R, Shorny S, Fassler R, Propst F, Wiche G: **Targeted inactivation of plectin reveals essential function in maintaining the integrity of skin, muscle, and heart cytoarchitecture.** *Genes Dev* 1997, **11**:3143-3156.

132. Sarkar PS, Appukuttan B, Han J, Ito Y, Ai C, Tsai W, Chai Y, Stout JT, Reddy S: **Heterozygous loss of Six5 in mice is sufficient to cause ocular cataracts.** *Nat Genet* 2000, **25**:110-114.

133. Klesert TR, Cho DH, Clark JI, Maylie J, Adelman J, Snider L, Yuen EC, Soriano P, Tapscott SJ: **Mice deficient in Six5 develop cataracts: implications for myotonic dystrophy.** *Nat Genet* 2000, **25**:105-109.