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Impaired Regeneration of Dystrophic Skeletal Myofibers Based on Ineffective Protein Utilization

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Abstract: A unifying concept in myofiber utilization of protein as a basis for muscular hypertrophy would implicate effects on viability of cells. Skeletal and cardiac myofibers would constitute post-mitotic cell phenotypes that developmentally are determined by structural organization and evolving participation of a homeostatic balance between production and effective utilization of specific protein moieties. Muscular dystrophies might be characterized as failed attempts at regeneration of myofibers subsequent to a variety of injuries based on sarcolemmal or other cell components including the nuclear membrane and sarcomere. Myofiber hypertrophy itself appears an abnormal attempt at compensation for a myofiber injury that involves also increase in intermediate filaments intracytoplasmically. Intermediate filament biology would strictly characterize not only the skeletal or cardiac myofiber, but also driving forces in the operative processes of hypertrophy and regeneration of the myofibers as post-mitotic highly differentiated cells.

Key words: Protein, cardiac, skeletal, utilization

INTRODUCTION

A myofiber appears to undergo a partial dedifferentiation in the process of regeneration after significant injury. The microscopic appearance of cytoplasmic basophilia together with internal nucleation as the injured myofiber undergoes a more rounded profile would be suggestive of regression to a fetal type of morphology. Other phenotypic attributes would also tend to promote any regenerative efforts on the part of the myofiber. Downregulation of a novel Regeneration-Associated Muscle Protease (RAMP) could be involved in progression of Duchenne muscular dystrophy[1]. Arrested maturation of myofibers (as suggested also with centronuclear congenital myopathy) and possible reversion to an immature phenotype would appear central to regenerative responses of skeletal myofibers.

Enhancement of regeneration would ideally ameliorate various disease processes that target not only skeletal muscle in dystrophies but also cardiac muscle that has undergone recent infarction. Heart failure from cardiomyopathy is a major cause of death in Duchenne dystrophy[2].

Smooth muscle appears capable of undergoing responsive alterations in terms of both hypertrophy and possibly also hyperplasia of myofibers. The vascular smooth muscle hypertrophy seen in systemic hypertension indicates a high degree of responsiveness in adaptation by hypertrophy and abortive hyperplasia.

The uterine hypertrophy of pregnancy occurs on a physiologic basis with a fundamental tendency to undergo various adaptive changes of the myofibers.

Skeletal myofibers in the muscular dystrophies appear less capable than normal to undergo regeneration. Limited regeneration may be strongly linked to the active dystrophic process itself. Sarcolemmal deficiency of dystrophin, as seen in Duchenne and Becker dystrophies, might be implicated. In Duchenne muscular dystrophy, utrophin may compensate for dystrophin during development[3]. This study appears substantiated by the often-occurring secondary deficiency of a series of other protein moieties that normally form part of the integral transmembrane complex with dystrophin. A primary deficiency of dystrophin would not only contribute significantly to active myofiber degeneration but also to a particularly poor regenerative capability once degeneration has become established.

Such defective regenerative activity might be central to a distinction between Duchenne muscular dystrophy and the Becker variety in terms of degree of clinical severity. Controlling the intracellular and extracellular microenvironment may promote an ideally maintained series of regenerative attempts at recovery of myofibers.

The active dystrophic process appears particularly characterized by an impaired regenerative process involving abnormal reorganization and failed maintenance of normal cytoarchitecture of the myofiber.

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Contraction- induced injury is placed on myofibrillar and membrane proteins[4].

The large hypertrophic dense myofibers characteristic of Duchenne muscular dystrophy would constitute active attempts at failed hypertrophy as a result of deficiency of dystrophin and other cytoskeletal proteins. A deficient cytoskeleton would prove insufficient in maintaining a structure promoting possible active hypertrophy and regeneration.

The degree of commitment of the cell subjected to normal physiologic circumstances of contractility would determine to some extent ability for myofiber regeneration. A cell such as the neuron or the myofiber is heavily committed to a specialized heavy production of intermediate filaments that appears implicated in possibly limiting regenerative response.

Abundant intermediate filaments involve not only a limitation on the various possible responses of a basic type, but a real characterization of the highly specialized differentiation of the myofiber as a normal contractile unit. The hypertrophied myofiber would be less prone to undergo regenerative activity due to an even heavier commitment to contractility and production of intermediate filaments.

A particularly vicious circle would involve skeletal myofibers in patients whose muscles are actively undergoing dystrophy. Cardiac myofibers in ventricular hypertrophy often develop to a large size after a sizeable left ventricular infarct.

Such unfavorable circumstances would inhibit maintained regenerative attempts at recovery of surviving myofibers. Such attempts at partial resolution of the issue of insufficiency in muscle mass would correlate also with decreased potential for compensation by injured myofibers.

A concept of limited commitment for regenerative response would go hand in hand with a contractility based on cytoskeletal dynamics that are often dependent on an integral sarcolemma.

Both myofiber hyperplasia and hypertrophy would appear to operate as a structural and pathobiologic continuum determining myofiber recoverability from injury.

A failed capability for vigorous myofiber hyperplasia may be linked to an ineffectively maintained hypertrophy of many myofibers injured as a result of processes such as dystrophy or ischemia.

The majority of boys with Duchenne muscular dystrophy carry a deletion in the dystrophin gene[5].

Duchenne muscular dystrophy appears a process of impaired response that spans multiple physiologic functions and that arises as a maintained insufficiency of hypertrophic and hyperplastic recovery of initially injured myofibers. Hematopoietic stem cell transplantation fails to restore experimentally dystrophin expression in Duchenne muscular dystrophy[6].

A structural reorganization of the myofiber would be necessary in terms of various processes that span regeneration, hypertrophy and hyperplasia to replace lost or injured myofibers.

The highly differentiated state of myofibers would not fully account for a weak series of regenerative attempts. The actual cytoskeletal apparatus with its peculiar stress-inducing effects on the sarcolemma and the heavy complement of intermediate filaments in the cytoplasm would render myofiber contractility an inhibitory influence on regenerative activity.

Actin and myosin filaments in particular appear to influence potential myofiber regeneration in terms especially of a myofiber mass that actively contracts under normal physiologic circumstances.

Dystroglycan is implicated in basement membrane formation linking dystrophin and laminin in muscle. It also modulates the actin cytoskeleton by associating with the cytoskeletal adaptor azrin[7].

Compensatory mechanisms as pathobiologic rather than as biologic systems: The phosphorylated state involving tau appears related to the active phosphorylating process as this evolves, rather than specifically to the formation of neurofibrillary tangles. The actual phosphorylating process would be directly incorporated within an abnormal evolution of tau microtubule associated fibrillogenesis. Both the phosphorylation of tau and the intrinsic process of neurofibrillary tangle formation would overshoot a basic framework of operability that constitutes pathologic involvement. It is in this sense that neurofibrillary tangles constitute a component of intermediate filaments normal to neuronal physiology.

Neurofibrillary tangles and the actin/myosin aggregates that develop with degeneration or myofiber hypertrophy would constitute different aspects of a single basic phenomenon that over-compensates attempts at cell recovery. Compensatory mechanisms of myofiber hypertrophy would appear an intrinsically pathobiologic process that impairs reactivity and regeneration of myofibers as injured cell components.

Duchenne muscular dystrophy beyond simple disruption of the sarcolemma: Little is known regarding the cascade events leading to myofiber degeneration in Duchenne muscular dystrophy[8].

The skeletal myofiber constitutes an integral unit requiring firm fixation of contractile intermediate filaments to the sarcolemma[9]. The sarcolemma appears a determining factor in controlling contractility of the cytoskeleton. A global concept of dysfunctionality[10] would paradoxically help characterize muscular dystrophy in terms of a whole myofiber that fails to hypertrophy and to effectively regenerate.

A myofiber appears to implicate a viability that is intrinsically linked to its contractility. A serious defect in myofiber contractility would directly lead to degeneration.

On the other hand, maintained near-normal contractility of the myofiber would help ensure a viability that persistently improves capability for hypertrophy.

Defects in the dystrophin fixation bolt or of the sarcoglycan complex would result in muscular dystrophy as a mechanically mediated disruption of the myofiber cytoarchitecture and sarcolemma. The very complexity of organization of the contractile myofiber apparatus would be suggestive of a disruption central to a progressive degeneration in muscular dystrophy. Also, such complexity would render an integral approach to myofiber dystrophy essential in attempted recovery of cells and contractility. Protein compensation in dystrophic myofibers may be possible: Laminin alpha1 chain reduces muscular dystrophy in laminin alpha2 chain deficient mice[11].

Out of frame mutations of the dystrophin gene are generally present in patients with Duchenne muscular dystrophy. Such abnormality would possibly go beyond simple deletions or point mutations in rendering the dystrophin gene critical in determined viability of the myofiber. It appears highly significant that out of frame mutations of the dystrophin gene may result in an essentially abnormal reorganization of the whole myofiber that also impairs subsequent embryonic determination in evolution of the myofiber cytoarchitecture.

Distinct signaling pathways are differentially activated in skeletal muscle in mdx mice[12].

A dystrophic myofiber appears one primarily abnormal as a result of abnormal organizational assembly of much of its constituent components making up the contractile apparatus. Relative interactions of intermediate filaments with the sarcolemma and basal lamina appear particularly implicated.

Understanding the actual process of evolving full establishment of the organized structure of the skeletal myofiber as this develops would help account for development of muscular dystrophy due to out of frame mutation of the dystrophin gene. Dystrophin-deficient muscles are more susceptible to stretch-induced muscle injury and recovery from injury is incomplete[13] .

Orderly development of the myofiber during fetal life would determine the full viability of a myofiber in terms especially of a highly differentiated postmitotic cell type[14].

Sarcomeric Protein Components Impair Contractility and Also Viability of Myofibers Due to Impaired Membrane-based Interactions: One particular characterizing feature of non-sarcolemmal muscular dystrophies appears to involve the mode of interaction of mutated proteins with other proteins in the nucleus or for example in the sarcomere itself.

Abnormal membranes other than the sarcolemma itself would cause myofiber degeneration particularly as an integral unit. Interactions of the nuclear membrane components with laminin and peripheral chromatin appear particularly implicated in Emery-Dreifuss muscular dystrophies and in Limb-Girdle muscular dystrophy IB.

The nuclear envelope lamina network has functions suggestive of a molecular shock absorber. Mutated lamins can either destabilize nuclear architecture or influence nuclear responses to mechanical signals[15].

Lack of emerin in X-linked Emery-Dreifuss muscular dystrophy disrupts transcription factors that bind emerin and also destabilizes nuclear envelope architecture by weakening a nuclear actin network[16].

Certain molecular interactions would prove significant as abnormally involved interactive molecules that involve membrane-associated or structural components of the sarcomeres.

Preservation of a certain degree of normal contractility of the myofiber, and interactive phenomena involving various molecular species between nucleus and cytoplasm would determine essential myofiber viability.

An integral dystrophic process may implicate often an enzymatic defect such as expanded trinucleotide CTG repeat in the DM protein kinase gene in myotonic dystrophy, and mutations of calpain 3 in limb girdle muscular atrophy 2A[17].

In the latter condition, there has been a recent shift from molecular genetics towards biochemical assay of defective protein[18].

Increased cytosolic calcium concentration leads to myofiber necrosis in Duchenne's and may implicate exercise-induced calcium increases and downstream muscle degeneration[19].

Interactive participation of structural disorganization with possible enzymatic defects would implicate a membrane-based disruption or involvement of the sarcomere in characterizing a primary form of muscular dystrophy that targets viability of the integral myofiber.

Nitric oxide system disruption may contribute to the

development of muscular dystrophy[20]. Blockers of the stretch-activated channel can minimize short-term damage in muscles of the mdx mouse[21] .

Abnormal molecular interactions would account also for conduction cardiac block[22] and the development of cardiomyopathy in patients with Emery-Dreifuss muscular dystrophy. There is marked decrease in Ca(2+) binding proteins calsequestrin and sarcalumenin in dystrophin deficient cardiac muscle[23].

Intermediate filament type as a determining feature in cell differentiation: Intermediate filaments would appear to characterize the nature of specific cell specialization as a universal process of adaptation and transformation in the biology of cell forms. The differentiation process would be reflected in modifications and transformation of intermediate filaments intracellularly. Determination of intermediate filament biology might actually help account for forces that drive cell specialization and tissue differentiation in terms of integral biology of cells such as skeletal myofibers and contractility. The protease calpain 3 is implicated in myofibrillogenesis and sarcomere remodeling[24].

Such phenomena as phosphorylation and degree of oxidative stress would act as basic pathogenetic mechanisms in inducing transforming events and in determination of basic cell processes such as myofiber hypertrophy.

A balance in production and effective utilization of specific proteins in myofibers: Functionally related proteins are involved in metabolism and energy generation, cytoskeletal reorganization and biogenesis, growth and differentiation, calcium homeostasis, and also the serine proteases family[8].

The concepts of minus-protein dystrophies and surplus-protein congenital myopathies would indicate a fundamental abnormality within myofibers that goes beyond simple decrease or increase in specific protein categories.

Myofibers can adapt considerably; expression of several protein isoforms can be induced by either stretch or long-term change of activity. It appears that disease progression in Duchenne's can be slowed with changes of muscle activity[25].

Such an integral phenomenon would implicate an essential homeostatic mechanism in the normal control of protein production in terms of the amount of a whole series of proteins produced in relative amount to ensure viability of the myofiber. Calcineurin is a key mediator of hypertrophy and targets utrophin and also myostatin, a negative regulator of muscle growth[26].

Simply ensuring a correct absolute amount of each individual protein within a skeletal myofiber appears insufficient to ensure the correct relative production of the protein type in terms of myofiber physiology. Editing of dystrophin mRNA by induction of exon skipping, using antisense oligonucleotides, has been proposed to generate dystrophin expression in Duchenne muscular dystrophy[27].

Production of a protein type would perhaps often influence production of other protein molecular species. Also, functional links exist between calcium signaling and cytoskeleton[28].

Hence, one would account for the so-called secondary or associated protein deficiencies seen with reduction of sarcoglycans in dystrophinopathics, or reduction in calpain in dysferlinopathy[29,30]. One might consider a whole series of homeostatic mechanisms that operate as physiologic utilization of specific protein types in association with other myofiber protein species. Developmental biology of the skeletal myofiber might, in a true sense, be centered on a phenomenon of closely linked production and effective utilization of specific myofiber protein species. In oculo-pharyngeal muscular dystrophy, mutant polyadenylate binding protein nuclear 1 gene would result in failed accurate production of mRNA necessary in remodeling and maintenance of myocytes[31].

Such a postulated homeostatic mechanism would be seriously disturbed with a primary deficiency of a protein such as dystrophin in Duchenne muscular dystrophy.

Furthermore, such a postulated mechanism of inter-related production and utilization of specific proteins such as utrophin would help account for some of the essential differences pathobiologically between Duchenne muscular dystrophy and Becker's type. In the latter disorder, there is a relative deficiency rather than an absolute absence of dystrophin in the affected myofibers. This would possibly be indicative of a fundamental abnormality that reflects a lesser degree of clinical severity in Becker's muscular dystrophy based on operative production of a whole series of protein species in production and effective utilization. An intragenic deletion/inversion in the Duchenne Muscular Dystrophy (DMD) gene may induce exon creation and result in a Becker muscular dystrophy phenotype[24].

Upregulation of integrin alpha7beta1 compensates for sarcoglycan in mediating cytoskeletal-membrane-extracellular matrix interaction[32].

Fukuyama-type congenital muscular dystrophy and congenital muscular dystrophy 1C commonly exhibit reduced glycosylation of alpha-dystroglycan in skeletal myofibers. Fukutin-related protein appears implicated in

o-mannosylglycan synthesis of alpha-dystroglycan[33]. Modulation of LARGE (glycosyltransferase) may bypass alpha-dystroglycan glycosylation in distinct congenital muscular dystrophies[34].

Congenital myopathies, on the other hand, as a single unified concept, would represent a dysregulation resulting in excess surplus of specific protein types leading to accumulation of proteins. A defective developmental series of pathways would account for a structural anomaly that impairs viability of myofibers.

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