

REVIEW

Gem Depletion: Amyotrophic Lateral Sclerosis and Spinal Muscular Atrophy Crossover

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SUMMARY

The determining factor of spinal muscular atrophy (SMA), the most common motor neuron degenerative disease of childhood, is the survival motor neuron (SMN) protein. SMN and its Gemin associates form a complex that is indispensable for the biogenesis of small nuclear ribonucleoproteins (snRNPs), which constitute the building blocks of spliceosomes. It is as yet unclear whether a decreased capacity of SMN in snRNP assembly, and, hence, transcriptome abnormalities, account for the specific neuromuscular phenotype in SMA. Across metazoa, the SMN-Gemins complex concentrates in multiple nuclear gems that frequently neighbour or overlap Cajal bodies. The number of gems has long been known to be a faithful indicator of SMN levels, which are linked to SMA severity. Intriguingly, a flurry of recent studies have revealed that depletion of this nuclear structure is also a signature feature of amyotrophic lateral sclerosis (ALS), the most common adult-onset motor neuron disease. This review discusses such a surprising crossover in addition to highlighting the most recent work on the intricate world of spliceosome building, which seems to be at the heart of motor neuron physiology and survival.

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Introduction

The correct synthesis, handling and processing of messenger RNAs (mRNAs) is fundamental for the manufacturing of functional proteins, without which, life in its most basic or complex form is simply impossible. Splicing or the pruning away of protein noncoding sequences (introns) and the fusion of coding segments (exons) is particularly important in transforming a pre-mRNA just transcribed from DNA into a mature mRNA that can be readily translated. The machine responsible for orchestrating this ongoing process is the spliceosome, which, at its core, is essentially a coalition of small nuclear ribonucleoproteins (snRNPs). The intricate world revolving around spliceosome building would have remained obscure had a catastrophic neurodegenerative disease not been discovered in which snRNP assembly was found to be “out of tune” [1–3]. Characterized by loss of lower motor neurons as well as progressive muscle weakness and wasting, spinal muscular atrophy (SMA) is a motor neuron disorder (MND) that is recessively inherited, and as there is currently no cure, it remains the most common genetic cause of infant death [4]. In the majority of cases (>97%), SMA has been pinned on low levels of the ubiquitously expressed survival motor neuron (SMN) protein as a result of homozygous deletions or, less frequently, mutations in the *SMN1* gene, and a lack of full compensation by a duplicate gene, *SMN2* [5]. Copy number polymorphisms of *SMN2* and,

thereby, SMN protein levels, are coupled with the degree of phenotype intensity in SMA [2,6,7].

Although the biology of the SMN protein has been intensely explored throughout more than two decades since its discovery as the factor determining SMA, three fundamental truths remain. First, SMN is oligomeric in nature [8–11] and stably associates with a set of core Gemin proteins to form the SMN-Gemins complex, the size and membership of which is dependent on organism complexity [12]. Second, SMN and its Gemin associates are indispensable for the biogenesis of snRNPs [13–16], and in case of SMN, a tight correlation exists between the degree of its deficiency and snRNP assembly capability [3,17,18]. Third, across metazoa, the SMN-Gemins complex concentrates in multiple nuclear puncta named gems or Gemini of Cajal bodies (CBs) in view of them frequently neighbouring or overlapping CBs [19,20]. The number of gems has long been known to be a faithful indicator of SMN levels and, hence, SMA severity [2,14,21]. Intriguingly, a flurry of recent studies have revealed that depletion of this nuclear structure, which is symptomatic of low SMN levels, is also a hallmark feature of the most common adult-onset MND, amyotrophic lateral sclerosis (ALS) [22–28]. These emerging findings raise the question of whether spliceosome building is at the heart of motor neuron physiology and survival. Should this be the case, therapeutics aimed at modulating this pathway most likely through augmentation of SMN levels may be universally beneficial in

MND. In this account, I review the most recent work on spliceosomal snRNP assembly including the surprising discovery that the less famed Gemin associates of SMN have a key stake in this process and, in this context, their loss of function *in vivo* disrupts normal motor function in a similar manner to reduced levels of SMN. Importantly, I highlight why SMN has been and, still remains more than ever, the best door to successfully gain insights into how building blocks of the spliceosome are manufactured and why this process might selectively impact the motor unit, when it gets “out of key”.

Recent Developments in Spliceosome Building

Molecular Architecture of the snRNP Assemblyosome

In vertebrates and, hence, humans, the SMN-Gemins complex is an elaborate nine-membered union of diverse proteins that largely do not share sequences or domains (Figure 1). SMN, seven Gemin proteins (Gemin2–Gemin8) [16,29–36], and Unrip [37,38] complete its membership. Not all metazoan organisms are endowed with a sizable complex, thus the fruit fly *Drosophila melanogaster* possesses a minimalistic SMN-Gemins complex counting only SMN, Gemin2, Gemin3 and Gemin5 amongst its members [12,39–42]. In addition to suggesting that the SMN-Gemins complex gained complexity in evolution, the simplicity of the fly version is an alluring feature considering that *Drosophila* has a well-earned reputation as a superb model organism of neurodegenerative disease (reviewed in [43]). Although they come together to establish the snRNP assemblyosome, SMN-Gemins complex members have additional unrelated functions and select components including Gemin3 and Gemin5 even participate in alternative multi-subunit complexes with a function in an snRNP-independent activity that is nonetheless RNP-centric (reviewed [12]).

The protein–protein interactions within the vertebrate SMN-Gemins complex are quite intricate, however, a consensus interaction map was recently drafted that establishes a troika formed of SMN, Gemin2 and Gemin8 at the backbone of the human SMN-Gemins complex that is capable of recruiting the remaining members in blocks. In this respect, SMN associates to Gemin2, Gemin3, and Gemin8. Gemin2 and Gemin3, in turn, interact with Gemin5 and Gemin4, respectively, and Gemin8 interacts with Gemins 4 and 7 [11]. UNRIP and Gemin6 are recruited by Gemin7, which is also capable of associating with Gemin2 [11,44]. Interestingly, in addition to SMN, only Gemin2 and Gemin8 were reported to self-associate [9–11,45] with the Gemin2–Gemin2 interaction stabilizing SMN oligomers [45]. Furthermore, Gemin5 and UNRIP are viewed as partial rather than full members in view of their weak and cytoplasmic-restricted association with the human SMN-Gemins complex [11,37,38,46,47].

Leading Roles for the Gemins in snRNP Assembly

snRNPs consist of one or two short noncoding RNA molecules (small nuclear RNAs or snRNAs) bound to a set of seven

Smith (Sm) or Sm-like (Lsm) proteins, and a unique set of snRNP-specific proteins [48]. In partnership with numerous non-snRNP splicing factors, U1, U2, U4/U6 and U5 snRNPs form the major spliceosome, which is responsible for splicing the majority of pre-mRNA introns, whereas U11, U12, U4atac/U6atac and U5 snRNPs constitute the less abundant minor spliceosome, that is specialized in processing a rare group of introns [49]. In contrast to Lsm-class U6 and U6atac snRNPs, which are synthesized entirely in the nucleus [50], for reasons as yet unclear, the core structure of the remaining Sm-class snRNPs is assembled in the cytoplasm. Several studies of late have only scratched the surface of the intricacies, intrigues and vicissitudes defining this key segmented phase *in vivo*, which is ATP-dependent, requires the participation of the entire SMN-Gemins complex [11,13–16,44,45], and essentially involves the uploading of a heptameric Sm D1/D2/E/F/G/D3/B ring-shaped core domain onto the “Sm site”, a conserved uridine-rich sequence motif intrinsic to snRNAs.

snRNAs that are expelled from the nucleus following transcription are thought to be swiftly identified by SMN-Gemins complex-independent Gemin5 protein through the stringent recognition of a code formed of sequences and structural motifs [15]. Gemin5 charged with snRNAs via its N-terminal WD-repeat domain [51], then docks into the SMN-Gemins complex, most probably proximate to Gemin2, to deliver its cargo for Sm core assembly [52]. The majority of Sm proteins are recognized by Gemin2, which wraps itself around a crescent-shaped pentamer formed of Sm D1/D2/E/F/G to contact each of the five Sm proteins. Importantly, the N-terminal tail of Gemin2 reaches into the snRNA-binding pocket on the Sm pentamer to block their inclination for promiscuous RNA binding, presumably until the delivery of the bona fide RNA substrates, snRNAs [53] (Figure 2). Gemins 6 and 7 form a heterodimer that resembles Sm core protein dimers even though they both lack significant sequence similarity with Sm proteins. In this context, the Gemin6/Gemin7 dimer is thought to serve as a surrogate for the SmB-SmD3 dimer around which the Sm pentamer is arranged, eventually facilitating the pentamer’s association with the Gemin2 arm of the SMN-Gemins complex [54]. Unrip may be important for the exchange of the Gemin6/Gemin7 heterodimer by the Sm D3/B particle, which should ensure Sm ring closure within the SMN-Gemins complex in preparation for uploading onto snRNAs [44]. The chaperoning of RNA and, eventually, RNP molecules as well as ATP breakdown during the assembly reaction, are probably entrenched jobs of the DEAD-box RNA helicase Gemin3 [30,36,55] assisted by its probable cofactor, Gemin4 [31].

It is unexpected that SMN does not feature in the crucial steps of snRNP assembly. Does this imply that it is not so important as previously thought for this essential housekeeping activity? Or, in view of substantial evidence (reviewed in [56,57]), does SMN burden itself with a less-defined non-canonical function, likely linked to axonal mRNP trafficking? It is the opinion of many that SMN’s function in snRNP assembly is the most decisive of all SMN-Gemins complex members, as its oligomers probably form a sturdy scaffold on which the Gemin builders operate in several concurrent assembly reactions following “due diligence” of the assembly blocks. This activity might be supported by other SMN-Gemins complex components especially those such as Gemin8, which are known to have a propensity to oligomerize [11]. Although exten-

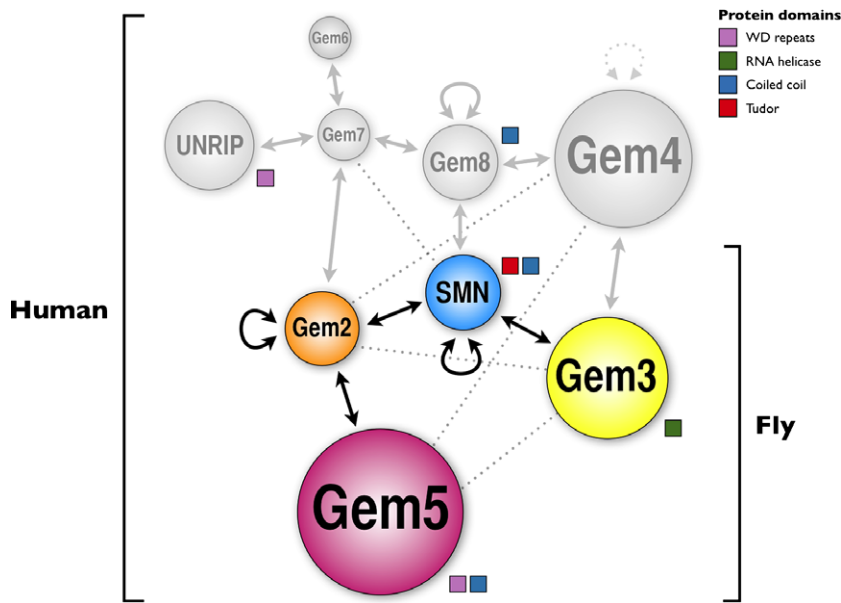


Figure 1 Survival motor neuron (SMN)-Gemins complex members: united in diversity. Interactions between SMN-Gemins complex members verified in one and more than one experimental system are depicted in solid and dashed arrows, respectively. Circular arrows indicate self-association. Protein domains in the Simple Modular Architecture Research Tool (SMART) database are also indicated for each member. Only the coiled coil domain is shared by more than one complex member. The fly version of the SMN-Gemins complex is minimalistic and composed of only SMN, Gemin2, Gemin3 and Gemin5. Addition of other components with evolution increased the complexity of the SMN-Gemins complex to the present nine-membered version present in vertebrates including humans (adapted from [12]).

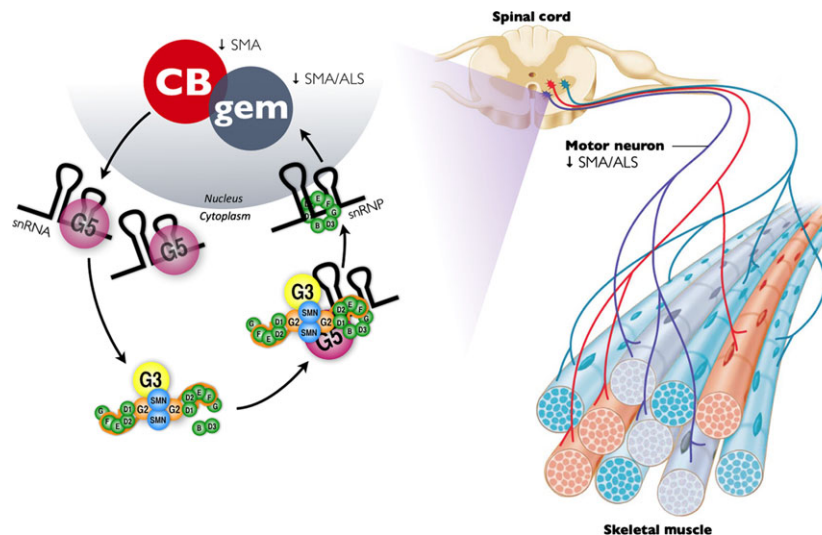


Figure 2 Cajal bodies, gems, spliceosome building and motor neuron degeneration. Newly transcribed snRNAs are exported to the cytoplasm where they are captured by Gemin5, and transferred to the survival motor neuron (SMN)-Gemins complex for assembly of a ring-shaped heptameric Sm protein core assembly. For simplicity, the SMN-Gemins complex depicted here is the *Drosophila* version. Gemin2 is the factor that recognizes and captures the majority of Sm proteins by wrapping itself around an Sm D1/D2/E/F/G pentamer. snRNP assembly is energetically demanding, and ATP hydrolysis is probably a function of DEAD-box RNA helicase Gemin3, which might also act as an RNA or RNP chaperone during the assembly reaction. Following Sm ring closure, snRNPs are imported into the nucleus, and prior to functioning in pre-mRNA splicing, they undergo maturation in the Cajal body (CB). The function of gems is as yet unclear but they associate with CBs and their loss is a signature marker of both spinal muscular atrophy (SMA) and amyotrophic lateral sclerosis (ALS) pathology.

sive *in vitro* studies reported on the possibility that the protein arginine methyltransferase 5 (PRMT5) complex sequesters and modifies select Sm proteins post-translationally, an action which is believed to enhance their interactions with the SMN-Gemins complex (reviewed in [58]), *in vivo* experiments concluded that this action is probably not indispensable for snRNP assembly [59]. Proper Sm core assembly is not only required for the *in vivo* stability and function of snRNPs but it is also a prerequisite for subse-

quent steps in the biogenesis of snRNPs including cap hypermethylation, 3' terminal trimming, and eventual import into the nucleus. In the latter compartment, snRNPs undergo maturation in CBs prior to their participation in pre-mRNA splicing including nucleotide modification guided by small CB-specific RNPs (scaRNPs), incorporation of several snRNP-specific proteins as well as coupling of individual snRNPs into di- and tri-snRNPs [12,60,61]. Following several rounds of action, snRNPs are known

to repeatedly return to CBs where they are regenerated or recycled [62].

The Link Between Spliceosome Building and Motor Function

Confirming their participation in a crucial housekeeping function, early lethality is the phenotypic endpoint of knockouts or strong global knockdowns of SMN-Gemins complex members analyzed thus far in a variety of model organisms [42,63–72]. However, SMA is the result of insufficient levels but not complete loss of the SMN protein, and replicating this situation in worm, fly, zebrafish and mouse gives rise to phenotypes that closely resemble SMA's signature pathology [5,41,63,65,73–77]. Considering their intimate liaison with SMN as well as their emerging key roles in spliceosome building, why are the Gemin genes not linked to SMA or similar motor conditions? The answer probably lies in the unique genetics of SMN in humans, hence, the absence of an *SMN2*-like pseudogene means that deleterious mutations in any of the Gemin genes is incompatible with life. Gemins (and Unrip) are also probably not redundant or interchangeable, therefore, they are responsible for exclusive activities during snRNP biogenesis. In this context, building on elegant biochemical and structural studies that support this direction [15,52,53], my laboratory demonstrated that organisms that have reduced levels of Gemins selectively in the motor unit develop motor deficits that are similar to those observed on attenuation of SMN [42,64]. In other words, if one considers the *in vivo* state, inadequate levels of any one member is sufficient to arrest the SMN-Gemins complex in a function that is critical for normal motor behaviour.

How can a decreased capacity of the SMN-Gemins complex account for the selective neuromuscular phenotype in SMA? Though still controversial, substantial evidence have been garnered to support one school of thought asserting that reduced snRNP biosynthesis and, consequently, defective splicing is selectively disruptive to the motor unit. To this end, early studies have demonstrated a significant phenotypic rescuing effect on restoration of normal snRNP levels in zebrafish and mouse SMA models [78,79]. More recently, in corroboration, studies on symptomatic severe SMA animal models uncovered an negative impact on splicing and expression levels of minor-intron-containing genes that are essential for motor-sensory circuit function [80,81], due to a previously described preferential reduction in the snRNPs that constitute the minor spliceosome [17,18,82]. The link between snRNP assembly and selective neuromuscular degeneration is, however, refuted by studies demonstrating that the role of SMN in snRNP assembly can be uncoupled from motor or axonal defects [83,84] and, recent reports have even presented evidence against a minor-intron-dependent aetiology for SMA [85,86]. Nevertheless, in a technically challenging study, which in contrast to others [18,87], involved the sequencing of RNA from microdissected motor neurons of presymptomatic SMA mice, the Dreyfuss' laboratory has yet again led the way by successfully identifying specific transcriptome abnormalities that link SMN deficiency to the distinct motor neuron pathology in SMA [86]. Overall, the evidence is substantial to tip the balance in favour of a vital role for SMN and, likely the SMN-Gemins complex, in preventing transcriptome irregularities.

MND Crossover

Gems and Cajal Bodies

What is the function of Gems? We are still in the dark when it comes to answering this question although great progress has been made on the Cajal body (CB), gems' twin nuclear structure. More than a century ago, making use of the silver staining technique, Spanish neuroscientist Santiago Ramón y Cajal described a nuclear structure that is generally smaller than nucleoli, but with which it is sometimes associated (hence, the original name *cuervo accesorio* or accessory body), in the neurons, including motor neurons, of many different vertebrates [88]. Although hosting an expanding list of proteins and RNAs or RNPs, the CB is usually identified by the presence of scaRNAs and its signature protein coilin (reviewed in [60,61]). CBs are the sites of several crucial steps in the maturation and regeneration of the RNA-processing machinery so it was quite surprising that loss of CBs through genetic depletion of coilin is not associated with lethality in *Drosophila* [89]. Furthermore, in this genetic background and in coilin- or SMN-deficient cells, scaRNA levels are normal and snRNAs are properly modified [90–92]. The story does not repeat itself in vertebrates, in that, besides the obvious CB dispersal phenotype [93], coilin knockout mice show reduced viability, fertility and fecundity [94], and in zebrafish, coilin depletion during embryogenesis leads to deficits in snRNP biogenesis, disrupted pre-mRNA splicing and consequently, reduced cell proliferation followed by developmental arrest [95]. These findings suggest that by concentrating several components in discrete nuclear compartments, CBs act as catalysts that accelerate reactions including those linked to snRNP biogenesis and this is especially important in highly metabolically active cells [61,96].

Gems are snRNP-free concentrates of SMN-Gemins complexes minus Unrip that in their majority occur as distinct nuclear structures in foetal tissues but increasingly colocalize with CBs in later developmental stages and adulthood [97,98]. Coilin, which binds to SMN, has been reported to mediate the recruitment of SMN-Gemins complexes, and, hence, gems, to CBs [99]. It is highly likely that gems are storage depots for supplementary SMN-Gemins complexes that are recruited to CBs most probably for the regeneration of worn snRNPs and, in agreement, the upregulation of either SMN or its Gemin associates is known to induce gem formation or promote an increase in gem numbers [20,26,42,47,100]. In addition, whereas canonical CBs and gems are lost on loss of SMN or Gemins, knockdown of snRNA export or cap modification factors only disrupted CBs [13,101–104], hence, dissociating gem stability from that of CBs as well as indicating that CB integrity is dependent on ongoing snRNP biogenesis (Figure 2). Truth must be told. Gems are not the only membrane-free cellular structures in which the SMN-Gemins complex concentrates, hence, in the cytoplasmic compartment, it accumulates with snRNPs and nuclear import factors in U bodies [40,105,106], which probably are the factories where the actual assembly reactions take place. In cultured neurons, the SMN-Gemins complex also localizes to large stationary and small actively transported snRNP-independent granules in neuronal processes, possibly underscoring a tissue-specific role in mRNP trafficking [107–110]. Similar to gems, crosstalk with compartment-specific

organelles is also observed for the cytoplasmic SMN-Gemins complex-rich bodies [40,103–105], and both nuclear and cytoplasmic bodies respond to metabolic changes in the cell [19,111].

Underpinnings of Gem Depletion in ALS

In both ALS and SMA, lower motor neurons in the anterior horn of the spinal cord are exceedingly vulnerable to injury, raising the question of whether both disorders share a common pathogenesis. ALS, which also affects upper motor neurons in the cerebral cortex, is mostly sporadic and, in this respect, only a minority of cases (5–10%) are inherited. Until recently, the field was fixated on the pathogenic processes resulting from dominant mutations in *copper/zinc superoxide dismutase 1 (SOD1)* gene, which account for ~20% of the familial ALS forms. These mutations appear to confer a toxic gain of function to the protein rather than loss of the metalloenzymatic activity that converts the highly toxic superoxide anion to molecular oxygen and hydrogen peroxide. A game changer was the identification of mutations in an additional diverse number of genes, including a subset that is intricately linked to RNA processing. Amongst these, RNA-binding proteins TAR DNA-binding protein 43 (TDP-43), and fused in sarcoma (FUS) are both nuclear proteins and shuttle between the nuclear and the cytoplasmic compartment as part of their cellular activities. Furthermore, under pathogenic conditions, both proteins are missing from their normal nuclear location and found bundled in cytoplasmic inclusions found in both neuronal and glial cells. This behaviour suggests that loss of normal nuclear function or, alternatively, gain of toxic function by aggregates play significant roles in ALS pathogenesis [112–115].

In the midst of the experimental frenzy surrounding the characterization of newly identified ALS-linked genes, Turner and co-workers were the first to audaciously probe for evidence favouring an involvement of SMN in the pathogenesis of ALS. To this end, although they found that TDP-43 distribution, expression or biochemistry was unaltered in spinal cords of affected SMA mice [116], transgenic mutant SOD1 mice were characterized by low SMN (and Gemin2) protein levels, but not mRNA, and a further reduction was found to accelerate the phenotypic severity associated with this model [117]. Importantly, replicating the murine situation, SMN protein levels are also significantly reduced (~50%) in the spinal cords of sporadic ALS patients [26], in agreement with earlier evidence demonstrating an association between lower *SMN2* copy numbers and increased severity of sporadic ALS [118]. Loss of total SMN protein, reflected in the depletion of motor neuron gems initiates before disease onset and is progressive, linked to the evolution of the disease [27,28]. Interestingly, despite the loss of gems, CB numbers are preserved in motor neurons of mutant SOD1 mice [27,117]. This phenotype indicates that mutant SOD1 interferes with the shuttling of the SMN-Gemins complex to CBs in motor neurons, at least in part, by disrupting the interaction between SMN and coilin. In this regard, SMN and its associate, Gemin2, were found depleted from the nucleus and enriched within the cytoplasm, a behaviour that is also followed by both endogenous and mutant SOD1 [26,28,117].

This mechanism underpinning gem depletion is cell-autonomous as mutant SOD1-expressing astrocytes, which are known to influence the neurodegenerative process (reviewed in [113]), do

not contribute to loss of gems in wild-type motor neurons [27]. Recently, in the reverse experiment, both the Turner and Monani labs have showed that neuronal overexpression of SMN, and, hence, elevated gem numbers, significantly improved neuromuscular function as well as motor neuron survival in mutant SOD1 mice [26,27]. Furthermore, upregulated SMN protein levels in spinal motor neurons conferred greater resistance to the degenerative effects of mutant SOD1-expressing astrocytes [27], reaffirming SMN's neuroprotective facet. It is noteworthy that overexpression of wild-type SOD1 protein does not alter SMN levels or ameliorate the neuromuscular and survival phenotype in a severe SMA model, an observation suggesting that SMN functions downstream of SOD1, and in this respect, phenotype modulation can only be one-way [117].

The plot got really exciting when the total number of gems was found to be altered in spinal motor neurons of mice by knockout of endogenous TDP-43 or overexpression of human TDP-43 [22]. Strikingly still, gem numbers are dramatically reduced in cells that are either transfected with mutant TDP-43 or FUS, are derived from patients with ALS or have disrupted levels of either protein [23–25]. Despite normal levels of SMN and Gemins on FUS (but not TDP-43 [25]) knockdown, gems were efficiently restored on upregulation of SMN. This result indicates that the requirement for FUS in gem formation is not due to an effect on protein levels of SMN-Gemins complex members, and similar to SOD1, SMN seems to act downstream of FUS [23]. TDP-43 is known to associate directly with FUS [119], and the latter acts downstream of TDP-43 in a pathway essential for normal survival and motor function [120,121]. Both TDP-43 and FUS were also reported to localize to gems [24]. Additionally, biochemical experiments reveal that both TDP-43 and FUS interact with the SMN-Gemins complex [23,24,122,123]. In case of FUS, the association with SMN is mediated through not only a direct binding between the two but also U1 snRNP, a finding that might hint at a form of collaboration on spliceosome building [23].

Downstream Consequences: snRNP Disruptions and Axonal Defects

Considering that the locale and/or levels of the SMN-Gemins complex are disturbed in ALS, one begs the question of how this scenario influences downstream pathways including snRNP biogenesis. In this regard, studies have provided conflicting evidence. Tsuiji et al. [24] report that snRNA levels are up-regulated in cells depleted of TDP-43. This phenotype is also present in spinal cords of sporadic ALS patients, where snRNPs were shown to accumulate extensively in motor neuron nuclei. However, findings by Ishihara et al. [25] are indicative of an alteration in the snRNA repertoire of TDP-43-depleted cells or symptomatic ALS tissues, a phenotype that is reminiscent of that reported in symptomatic SMA mice [17,18]. Furthermore, in a recent study, cytoplasmic mis-localization of FUS, in cells expressing mutant FUS, was found to cause partial accumulation of snRNPs to the cytoplasm [122]. Interestingly, crossover does not seem to be limited to processes underpinning spliceosome building but also extends to noncanonical functions associated with SMN, especially those confined to neuronal processes, and in this regard, it is well known that paucity of SMN gives rise to axonal defects (reviewed in [56]). In this

regard, FUS mutants were found to disturb the axonal localization of SMN. Importantly, overexpression of SMN rescued the axonal defects induced by mutant FUS, raising the question of whether such defects arise as a result of SMN sequestration by mutant FUS [123]. Further research on this theme is warranted and can potentially give a clearer picture.

Conclusion

It is presently conceivable to deduce that RNA metabolism is crucial to motor neuron physiology and survival. Clues have been exposed in a melange of studies including those that report the identification of MND-linked mutations in genes encoding proteins with crucial roles in RNA processing and the in-depth investigations that they triggered [124–126]. Combine these with studies that link splicing factors or snRNPs to neurodegeneration ([127,128]). The reason for the selective vulnerability of motor neurons is still opaque though it is worth noting that accumulating evidence is suggesting that MND is more far-reaching and, hence, it may not be a cell-autonomous disease of motor neurons (reviewed in [129,130]). Gems are quite prominent in motor neurons [2,21,97,98], and though they are probably not essential for cell survival given their absence in certain cell types [97], is it plausible that they work in conjunction with CBs to expedite the final steps of the snRNP production line and/or the regeneration of snRNPs? Investigations that address this open question can potentially impact our understanding of the molecular nature underpinning motor neuron degeneration.

Aiming at identifying compounds that augment cellular SMN, gem counts have already been successfully exploited either as an

integral component of image-based screens [131] or as a confirmatory assay [100,132]. The unexpected finding that gem depletion is a signature feature of ALS pathology, in addition to SMA (Figure 2), consolidates the use of this cellular phenotype to screen for novel pharmacologic modifiers that are universally applicable in MND. Importantly, ALS patients might also benefit from the various SMA-directed therapeutics in the pipeline. Principally relevant are those that boost SMN protein levels including viral and non-viral mediated gene delivery, oligonucleotide-mediated *SMN2*-directed splicing modification as well as small molecules that spur SMN expression (reviewed in [133]). The coming together of two major motor neuron degenerative disorders heralds a bright outlook for treatment strategies targeting their eradication.

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Conflict of Interest

The author declares no conflict of interest.

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