Mol Neurobiol (2007) 35:288–297
DOI 10.1007/s12035-007-0026-x

Laminins in Peripheral Nerve Development and Muscular Dystrophy

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Received: 14 September 2006 / Accepted: 28 November 2006 / Published online: 6 July 2007

Abstract Laminins are extracellular matrix (ECM) proteins that play an important role in cellular function and tissue morphogenesis. In the peripheral nervous system (PNS), laminins are expressed in Schwann cells and participate in their development. Mutations in laminin subunits expressed in the PNS and in skeleton muscle may cause peripheral neuropathies and muscular dystrophy in both humans and mice. Recent studies using gene knockout technology, such as cell-type specific gene targeting techniques, revealed that laminins and their receptors mediate Schwann cell and axon interactions. Schwann cells with disrupted laminin expression exhibit impaired proliferation and differentiation and also undergo apoptosis. In this review, we focus on the potential molecular mechanisms by which laminins participate in the development of Schwann cells.

Keyword Radial sorting · Schwann cells · Proliferation · Apoptosis · Differentiation · Integrin · Myelination

Introduction

Laminins, heterotrimeric molecules, are critical components of the ECM. At present, 5 α chains, 4 β chains, and 3 γ chains have been identified, and of the 60 potential trimeric molecules that could be generated from these chains, 15 have been observed [1–3]. Laminins are present in both central and peripheral nervous systems [2, 4–11], including the neuromuscular junction [12–17]. Laminin function in the nervous system is varied: they play roles in neurite outgrowth and axon pathfinding [18], brain development [1, 19, 20], pathology [21], and in peripheral nerves development [22–25]. Laminins play at least three overlapping roles in mammals [26]: (1) they compose a major structural element of the basement membrane [27, 28]; (2) they provide attachment sites for cells via cell surface proteins (e.g., dystroglycan) [29]; and (3) they act as ligands for receptors on cells (e.g., integrins) and thereby initiate signals that influence cell behavior and survival [30].

Among the 15 known laminin isoforms, laminin 2 (α2β1γ1), laminin 10 (α5β1γ1), and low levels of laminin 8 (α4β1γ1) are expressed in the endoneurium of the peripheral nerves [17, 31]. Laminins play critical roles in peripheral nerve myelination as α2 laminin gene mutations in mice (dy/dy and dy2J/dy2J) [32, 33] or humans (CMD) [34] result in muscular dystrophy and peripheral nerve myelination defects [35–37]. In the past few years, considerable progress has been made in understanding the function of laminins in PNS development and the mechanism by which laminins regulate Schwann cell behavior and survival. Therefore, in this review, we discuss the potential molecular mechanisms of laminins in PNS development.

Laminin Function in Peripheral Nerve Development

The importance of laminins in PNS development was identified when it was discovered that mutations in the α2 laminin gene caused peripheral neuropathies in both humans [34] and mice [32, 33, 38]. Mutant mice display

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axonal hypomyelination in which axon bundles are naked as they lack ensheathment and myelination [35, 39]. Furthermore, the endoneurium basal lamina is disrupted, nerve conduction velocity is reduced [40], and nodes of Ranvier undergo morphological changes [41].

Further studies revealed that peripheral nerve defects in laminin mutant mice or human patients are because of the abnormal development of Schwann cells that lack normal laminin expression. In the peripheral nerves of the α2 laminin mutant mice (dy and dy2J), both laminin α4 and laminin α1 are upregulated in the endoneurium, yet the phenotype is mild [17, 42] (Table 1). However, the Schwann cell-specific knockout of laminin γ1 chain abolished all laminin subunit expression in these cells [23, 25]. These mice exhibited severe hind limb paralysis, muscular dystrophy, and impaired survival with few mutant mice reach adulthood. The peripheral nerves are much thinner than controls and are grossly translucent rather than the opaque white of the normal myelinated nerves. The nerve conduction velocity is dramatically decreased. Under light or electron microscope, the nerve fibers are severely hypomyelinated. Most axons are naked and tightly compacted together, whereas Schwann cell numbers are dramatically decreased [25]. There are a few axons that contain myelin sheaths, which could be caused by the close contact of these Schwann cells to the perineurium or blood vessels that express laminins. During development, Schwann cells lacking laminin expression do not extend processes between axons and lack a continuous basal lamina [23, 25]. The induction of both Krox-20 and Oct-6 is not affected in the mutant Schwann cells, but the maintenance of Krox-20 and the downregulation of Oct-6 in later postnatal stages is impaired. The mutant Schwann cells are arrested at the premyelinating stage. During later embryonic and perinatal stages, Schwann cell proliferation is dramatically impaired. Postnatally, γ1 laminin-null Schwann cells exhibit increased apoptosis. Therefore, laminins play important roles in Schwann cell proliferation, differentiation, and survival during PNS development [25].

Mechanism of Laminin Function in the Regulation of Schwann Cell Proliferation

During embryonic development (mice at E12/E13), Schwann cell precursors are derived from the neural crest, migrate along the peripheral axons, and ultimately differentiate into myelinating or nonmyelinating Schwann cells. During late embryonic and perinatal stages, Schwann cells and their precursors proliferate vigorously to rearrange, sort, and ensheath axons [43]. In laminin γ1 knockout and dy2J/α4 knockout mice, Schwann cell proliferation is dramatically decreased during late embryonic and perinatal stages (Fig. 1). As axons are a

Table 1 Relationship between laminin chain expression and PNS phenotypes in laminin mutant mice

Laminin chain	Wild type		dy or dy2J		α4 KO		dy2J/α4 KO		γ1 KO	
	Nerves	Roots	Nerve+ sciatic nerves are partially affected, brachial nerves are nearly normal	Root +++ spinal root and cranial nerves are severely affected	Nerve+ similar to dy, dy2J but show polyaxonal myelination, and brachial nerves are nearly normal	Root± almost normal	Nerve+++ severe hypomyelination in all forelimb and hind limb axons	Root± completely sorted and myelinated without basal lamina	Nerve+++ severely hypomyelinated	Root++ severely hypomyelinated
α1	–	–	++	–	ND	ND	++	ND	–	–
α2	+++	+++	+	+	+++	+++	+	+	–	–
α4	+	+	++	+	–	–	–	–	–	–
α5	±	+	±	+	ND	ND	ND	ND	ND	ND
β1	+++	+++	+++	+++	+++	+++	ND	ND	–	–
γ1	+++	+++	+++	+++	+++	+++	ND	ND	–	–

ND: not determined.
References: [23, 24, 25] and [42]

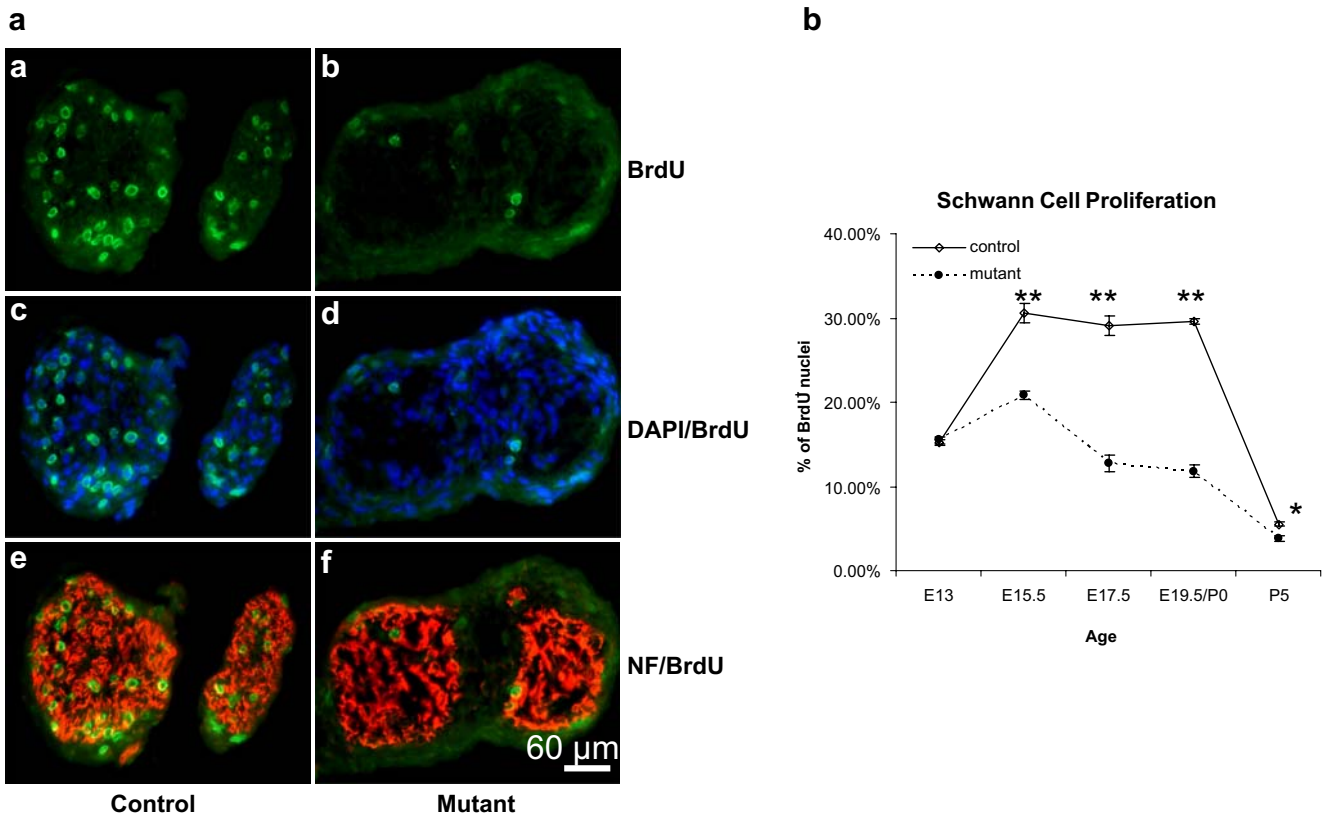

Fig. 1 Schwann cell proliferation is reduced in γ1 laminin knockout mice. **a** Transverse sections of sciatic nerve from control and mutant mice at E18.5 were triple stained for BrdU (*green*), DAPI (*blue*), and neurofilament (*red*). The images of BrdU/DAPI and BrdU/neurofilament (*NF*) were merged. Schwann cells in the mutant mice showed reduced nuclear BrdU incorporation (compare *b*, *d*, and *f* to *a*, *c*, and *e*). **b** Plot of the percentage of BrdU-positive nuclei at various developmental stages. The ratio of BrdU-incorporated nuclei are significantly reduced in mutant nerves at E15.5, E17.5, and E19.5/P0 ($n=6$ per genotype per day, $^{**}p<0.001$) (For materials and methods, refer to reference [25])

major source of Schwann cell mitogens [44, 45], impaired radial sorting can prevent Schwann cell exposure to axons, resulting in reduced Schwann cell proliferation. Neuregulin-β1 is a major axon-derived Schwann cell mitogen, which can interact with and trigger the phosphorylation of receptor tyrosine kinases ErbB2 and ErbB3 on Schwann cells [45]. In the peripheral nerves of γ1 laminin knockout mice, the levels of total ErbB2 and ErbB3 are similar to that of the control mice. However, the phosphorylation of these two receptors is dramatically decreased in the nerves of mutant mice [25]. Therefore, the inability of laminin-null Schwann cells to be exposed to axon-derived mitogens is a major cause of impaired proliferation in the γ1 laminin knockout mice. This result suggests that laminin-mediated interactions between Schwann cells and axons play an essential role in proliferation. Schwann cell proliferation in the peripheral nerves of dy2J/α4 knockout mice is also impaired; however, it remains unclear whether the phosphorylation of ErbB2 and ErbB3 receptors in these mice is also impaired [24]. As laminins can directly promote Schwann cell proliferation in vitro [24, 46, 47], it is possible that the lack of laminin expression also directly contributes to decreased proliferation (Fig. 4).

Laminin receptors involved in peripheral nerve development are α6β1integrin, α6β4, and dystroglycan [48]. Before birth, only α6β1integrin is expressed in Schwann cells [42]. However, Schwann cell proliferation is unaffected in the peripheral nerves of Schwann cell-specific β1integrin knockout mice [49]. This observation implies that the receptor mediating laminin function in Schwann cell proliferation during later embryonic stages is not understood. One hypothesis was that other laminin receptors, such as β4 or dystroglycan, are upregulated and compensate for the lack of β1. However, this is not the case, as neither β4 nor dystroglycan is upregulated in the peripheral nerves of β1 integrin knockout mice. Furthermore, other α chains that can normally form heterodimers with β1 integrin such as α2, α3, and α7 are also not upregulated [42]. These results raise the possibility that an unknown, alternative receptor may mediate laminin function in Schwann cell proliferation.

Mechanism of Laminin Function in Schwann Cell Survival

Throughout development, the mechanisms involved in Schwann cell survival are varied. At E12-13, the survival of Schwann cell precursors depends on axon-derived signals such as neuregulins/ErbB, whereas after E15, immature Schwann cells lose this dependency by establishing an autocrine signal to survive on their own [50]. As ECM proteins such as laminins play important roles in maintaining cell viability [8, 51], it is not surprising that Schwann cells lacking laminin expression exhibit increased apoptosis (Fig. 2). However, increased Schwann cell apoptosis in the γ1 laminin knockout mice only occurs during postnatal stages. At E15.5 and E17.5, there are very few apoptotic cells in the peripheral nerves and there is no difference between control and knockout mice. In contrast, the percentage of apoptotic cells in mutant nerves progressively increases in postnatal stages, peaking at P15, and then gradually declines as the nerves mature. Control nerves show very few apoptotic cells during these stages (Fig. 2).

As axon-derived survival signals appear to be only important in early postnatal developmental stages [52], it is unlikely that late postnatal cell death is caused by an improper Schwann cell/axon relationship. In the *P_0/Cre:fLAM γ1* knockout mouse line [25], laminin expression is disrupted around E13.5 to 14.5, and Schwann cell death increased at P0. In another γ1 laminin knockout mouse line, *CaMKII/Cre:fLAM γ1* [23], laminin expression in Schwann cells is ablated around E17.5, and the disruption is incomplete. Schwann cell apoptosis increased around P10 in the *CaMKII/Cre:fLAM γ1* model and is less severe than the *P_0/Cre:fLAM γ1* mice [25]. This delayed apoptosis suggests that laminins may be important for long-term survival of Schwann cells [53].

Several signaling pathways are important in regulating Schwann cell apoptosis, including PI 3-kinase/Akt and transforming growth factor β (TGFβ) pathways. In a cell culture system, PI 3-kinase activity is important for maintenance of Schwann cell viability [54]. TGFβ has been shown to induce Schwann cell embryonic apoptosis during development by activating c-*Jun* [55, 56]. In differentiating Schwann cells, expression of Krox-20 turns off TGFβ signaling and renders the Schwann cells resistant to apoptosis [57]. Schwann cells lacking laminin γ1 fail to express Krox-20 [23] at later postnatal stages. Therefore, lack of laminins in Schwann cells could result in an activated TGFβ death signal and a reduced PI 3-kinase survival signal, rendering the Schwann cells prone to apoptosis. In determining the signaling pathways involved in Schwann cell apoptosis in the laminin γ1 knockout mice, the phosphorylation levels of Akt/PKB protein, Jun-N-terminal kinase (JNK), and c-*Jun* (for TGFβ

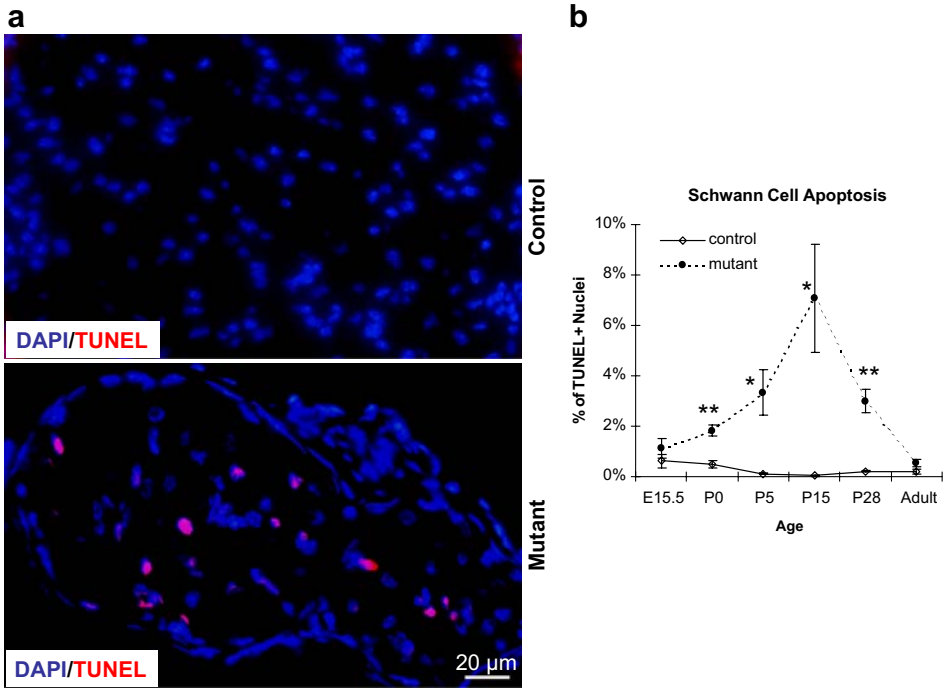

Fig. 2 Schwann cells in γ1 laminin knockout mice undergo apoptosis. **a** Transverse sections of sciatic nerve from control and mutant mice at P12 were stained with TUNEL (*red*) and DAPI (*blue*), and the images were merged. Apoptotic cells were obvious in the mutant nerve but not detected in the control nerves at this stage. **b** Plot of the percentage of TUNEL-positive nuclei at various developmental stages. The ratio of TUNEL-positive nuclei are significantly higher in mutant than in controls at P0, P5, P15, and P28 ($n=6$ per genotype per day, $^{**}p<0.001$) (For materials and methods, refer to reference [25])

signaling pathway) were analyzed. In the peripheral nerve of knockout mice, the phosphorylation levels of Akt at P0, P5, P15, and P28 were significantly lower than that of the control [25]. In adults, it is also slightly lower than the control. Further analysis revealed that the phosphorylation of glycogen synthase kinase 3β (GSK-3β) in the peripheral nerve of these knockout mice is also significantly lower than that of the control. Common markers of apoptosis, caspase-9, -3, and -7 were found in the activated form in the mutant nerves [25]. Differences in phosphorylation levels of JNK and c-*Jun* in peripheral nerves from mutant and control mice were not detected. Therefore, laminins may maintain Schwann cell viability through PI3K/Akt signaling pathway (Fig. 4).

In α2, α4, and dy2J/α4 laminin knockout mice, increased Schwann cell apoptosis was not observed [24]. However, in these mutant mouse lines, other laminin subunits were expressed. For example, in dy or dy2J mutant mice, both α1 and α4 are upregulated in peripheral nerves. In α4 knockout mice, α2 expression remains the same as control mice. In the dy2J/α4 knockout mice, α1 is significantly increased and a small amount of α2 chain is still expressed in the Schwann cells (see Table 1). These observations indicate that they may be functionally compensating for the lack of other laminin chains. In the laminin γ1 knockout mice, however, expression of all laminin subunits is abolished. These differences may explain why only laminin γ1 knockout mice exhibit increased Schwann cell apoptosis.

In the β1 integrin knockout mice, increased Schwann cell apoptosis is not observed [49]. As the lack of laminins cause increased Schwann cell apoptosis only after birth, at this stage, other laminin receptors such as β4 and dystroglycan are expressed and may compensate for the lack of β1 integrin. It will be interesting to see whether the β1 integrin/dystroglycan double knockout mice show increased Schwann cell apoptosis.

Mechanism of Laminin Function in Schwann Cell Differentiation

In the peripheral nerves of later postnatal and adult laminin γ1 knockout mice, there are only two types of Schwann cells: one type that forms myelin and still associates with laminins for unknown reasons, and another type that is arrested at a premyelinating stage, has a disrupted basal lamina, and does not have laminins (Fig. 3) [23, 25]. This phenotype indicates that laminins are important in Schwann cell differentiation.

Transcription factors that play key roles in Schwann cell differentiation are the POU domain protein Oct-6 (also known as Tst-1 or SCIP) and the zinc-finger protein Krox-20 [58–60]. Oct-6 is expressed in all Schwann cells in late embryogenesis and early postnatal periods with the highest

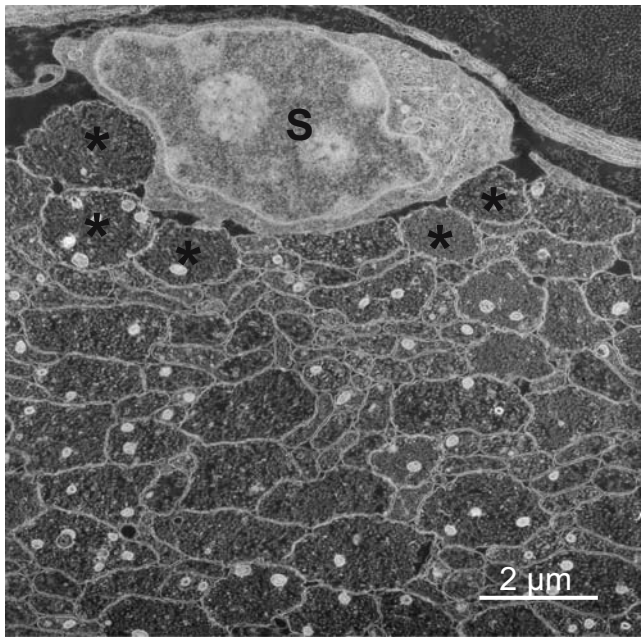

Fig. 3 Schwann cells lacking laminin expression are arrested at the premyelinating stage. An electron micrograph of P28 mutant sciatic nerve shows that a Schwann cell (*S*) that does not have continuous basal lamina was unable to extend cytoplasmic processes between axons (*asterisks*) and arrested at the premyelinating stage (For materials and methods, refer to reference [25])

expression in promyelinating Schwann cells [60, 61]. In contrast, Krox-20 is expressed only in the myelin-producing Schwann cells, which continue to express detectable levels of Krox-20 protein throughout life [62, 63]. Genetic and cell biological studies suggest that these transcription factors can interact with each other [63–67]. In promyelinating Schwann cells, Oct-6 is strongly induced by axonal contact-related signals (e.g. neuregulin I type III) [68], and Krox-20 is expressed subsequently. High expression of Krox-20 is required for the downregulation of Oct-6 after the peak of myelination and for the activation of the major myelin genes, such as P_0, myelin basic protein (MBP), and enzymes required for synthesis of normal myelin lipids. In laminin γ1 knockout mice, the induction of Oct-6 expression in Schwann cells is similar to that of control mice during late embryonic and early postnatal stages. During late postnatal stages, Schwann cells in control mice gradually downregulate Oct-6 expression, but Schwann cells in mutant mice failed to downregulate Oct-6. Induction of Krox-20 in mutant Schwann cells is similar to that of the control, but during later postnatal stages, the maintenance or upregulation of Krox-20 in mutant Schwann cells is impaired [25]. Therefore, laminins are not required for the induction of Oct-6 and Krox-20 but is instead essential for the maintenance of Krox-20 expression and the downregulation of Oct-6.

As axonal sorting is necessary for Schwann cell differentiation and this process is severely impaired in the

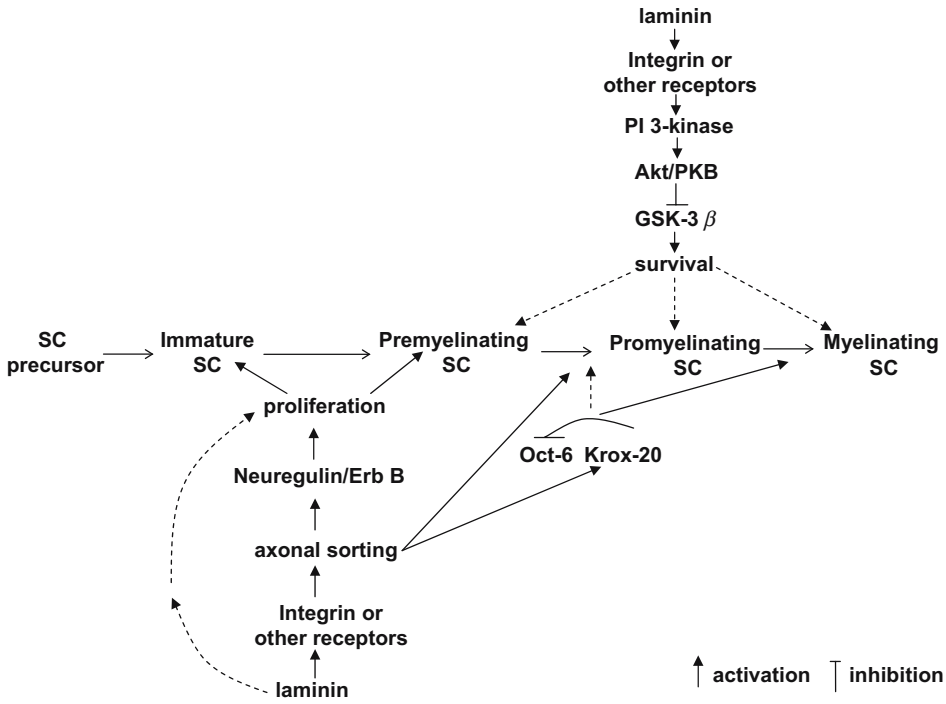

Fig. 4 A proposed model of laminin function in Schwann cell development (based on the results from references [25, 49]). Laminins mediate Schwann cell/axon interactions through integrin and/or other receptors, leading to axonal radial sorting. During this process, Schwann cells are exposed to axon-derived mitogens such as neuregulin. Neuregulin binding to Erb B receptors on Schwann cells triggers their proliferation (phosphorylation of Erb B receptors is reduced and Schwann cells proliferation is impaired in laminin KO mice). Laminin-mediated axonal sorting enables the transition of premyelinating Schwann cells to promyelinating Schwann cells and upregulates Krox-20 expression, which then downregulates Oct-6. Schwann cells lacking laminin expression are arrested at the premyelinating stage, Krox-20 expression is not sustained, and Oct-6 is not downregulated in postnatal stages in laminin mutant nerves. This regulation of gene expression further promotes myelinating Schwann cell differentiation and axonal myelination. Laminins also play important roles in Schwann cell survival. Laminins binding to integrin receptors activates PI-3 kinase, which phosphorylates and inactivates GSK-3 β. Inactivation of GSK-3 β suppresses apoptosis and therefore promotes Schwann cell survival. As there is a delay between loss of laminin expression and increased apoptosis, laminins may be required for long-term survival of Schwann cells

peripheral nerves of the laminin γ1 knockout mice, the changes in Oct-6 and Krox-20 expression in mutant Schwann cells may be a consequence of impaired radial sorting. In Oct-6 or Krox-20 knockout mice, axonal radial sorting is normal [59, 69], indicating that regulation of Oct-6 or Krox-20 expression is downstream of axonal radial sorting. We speculate that impaired regulation of Oct-6 or Krox-20 expression in the laminin γ1 mutant Schwann cells is because of the failure of laminin-null Schwann cells to sort axons (Fig. 4). We further deduce that incomplete axonal sorting also contributes to impaired Schwann cell differentiation (Fig. 4).

Laminin in Axonal Radial Sorting

Axonal radial sorting in the peripheral nerves of all laminin mutant mice is impaired [24, 25], and this impaired radial sorting may be the underlying mechanism for many of the phenotypes exhibited in these mutant mice. For example, reduced Schwann cell proliferation in laminin γ1 knockout mice is because of the incomplete radial sorting or inaccessibility of Schwann cells to axon-derived mitogens, and impaired Schwann cell differentiation could also result from improper axon sorting. How laminins regulate Schwann cell sorting of axons is currently unknown. As ECM proteins regulate cytoskeleton organization in many cell types [29, 70, 71] and Schwann cells lacking laminin expression fail to extend cytoplasmic processes, laminins may participate in Schwann cell sorting of axons by regulating cytoskeleton arrangement. Mice with a β1 integrin deficiency in their Schwann cells also exhibit impaired radial sorting of axons [49]. Therefore, β1 integrin-containing receptors may mediate laminin function in axonal sorting.

Focal adhesion kinase (FAK) and paxillin play critical roles in β1 integrin-dependent signaling [72, 73]. β1 integrin, FAK, and paxillin colocalize with each other at focal adhesions [74]. FAK is an intracellular protein tyrosine kinase that rapidly autophosphorylates in response

to β1 integrin binding to ECM [75]. Paxillin is an adaptor protein that integrates adhesion-dependent signals with changes in actin organization and gene expression. It is phosphorylated after cell activation by the ECM [73]. FAK and paxillin specifically coimmunoprecipitate with β1 integrin in differentiating SC/DRG neuronal cocultures but not in Schwann cell-only cultures, and tyrosine phosphorylation of FAK and paxillin increase as Schwann cells form basal lamina and differentiate [76]. In addition, actin has also been implicated in both changes in cell shape and gene expression associated with Schwann cell differentiation [77]. Therefore, it is possible that laminins may bind to β1 integrin and recruit FAK and paxillin as signal transducers to induce cytoskeleton reorganization for axonal sorting.

Role of Different Laminin Isoforms in PNS Development

Although the many versions of laminin mutant mice all show a similar hypomyelination phenotype in their peripheral nerves, there are some differences between these mice: (1) In the peripheral nerve of α4 mutant mice, there are some polyaxonally myelinated small bundles (multiple unsorted axons wrapped by myelin sheath), which is rare in α2 and γ1 laminin knockout mice; (2) the spinal root is nearly normal in α4 knockout mice, but hypomyelination is more severe in the root than in the distal part of peripheral nerves in α2 mutant mice. Spinal root and nerves are similarly affected in γ1 laminin knockout mice; (3) Basal lamina in the PNS is normal in α4 mutant mice, but is disrupted or discontinuous in α2 mutant and γ1 laminin knockout mice; (4) Schwann cells in γ1 laminin knockout mice undergo increased apoptosis, which is not observed in α2, α4, and dy2J/α4 double mutant mice [24, 25]. These differences may be a result of compensation from different isoforms (Table 1), suggesting that laminin isoforms may play distinct roles in different areas of the PNS during development.

In the distal part of peripheral nerves of dy and dy2J mice, both α1 and α4 are upregulated and the nerve only shows minor defects in axon sorting and myelination. In contrast, only α4 is expressed in the spinal root and myelination defects are more severe in this region. Therefore, laminin 1 (α1β1γ1) alone or incorporation with laminin 8 (α4β1γ1) promotes axon myelination in the distal part of peripheral nerves of dy or dy2J mice (see Table 1). In α4 knockout mice, expression of other laminin chains does not change, and peripheral nerves show minor defects whereas spinal roots are nearly normal, indicating that laminin 8 is necessary for myelination of some axons in the distal nerves. It remains unclear whether laminin 2 and 8 are respectively responsible for myelination of specific subpopulations of axons in the PNS. In dy2J/α4 knockout and α2/α4 double knockout mice, distal nerve myelination is severely affected. In the spinal roots of dy2J/α4 knockout, α5 and mutant α2 are expressed and axons are completely sorted and myelinated although there is no basal lamina formed, demonstrating that the basal lamina is not required for myelination. In α2/α4 double knockout mice, both root and distal nerves are severely hypomyelinated, which is similar to γ1 laminin knockout mice. Therefore, laminin 2 and 8 together are responsible for axonal sorting and myelination in both spinal root and peripheral nerves. However, in the spinal root of dy2J/ α4 knockout mice, mutant laminin 2 (α2) cooperates with laminin 10 (α5) and can sort and myelinate almost all the axons. In the spinal root of dy2J mice, laminin 8 (α4) somehow interferes with the collaboration between mutant laminin 2 and laminin 10; therefore, axons cannot be sorted and myelinated. When laminin 10 is ectopically expressed in the peripheral nerves of dy2J/ α4 knockout mice, axonal sorting and myelination is dramatically increased, indicating that laminin 10 promotes axonal sorting and myelination through collaboration with mutant laminin 2 [24]. In γ1 laminin knockout mice, where compensation by other laminins is not possible, the hypomyelination phenotype is most severe [25] (Table 1).

Laminins in Muscular Dystrophy

There are three spontaneous α2 laminin mutant mouse lines that cause dystrophy (for review see [41]). They are Lama2^{dy} [78], Lama2^{dy2J} [79], and Lam^{dypas} [80]. As knockout of α2 laminin in mice showed similar muscular dystrophy and peripheral neuropathy to *dystrophic* mice [81–83], these mutations most likely cause loss-of-function of α2 laminin. In *dystrophic* mice, both muscle and peripheral nerves are affected, and the phenotypes are a combination of nerve and muscular pathology. How peripheral neuropathy contributes to the phenotype is not clear. A recent study showed that muscle-specific expression of a human α2 laminin transgene in α2-deficient mice greatly improved muscle pathology. However, these animals still exhibit progressive lameness of their hind legs, which may be caused by the uncured peripheral nerve hypomyelination [82]. This study suggests that peripheral neuropathy is a critical pathologic component of the dystrophic phenotype in these mutant mouse lines. Our studies from Schwann cell-specific γ1 laminin knockout mice also demonstrate that laminin deficiency in peripheral nerves alone without affecting skeleton muscle can cause severe muscular dystrophy and hind limb paralysis [23, 25].

Mutations in α2 laminin cause merosin-deficient congenital muscular dystrophy in humans [34] (MD-CMD or

MDC1A), which is the most common type of congenital muscular dystrophy. These mutations cause deficient or nonfunctional laminin 2. The pathologic changes in the peripheral nerves of MD-CMD patients are similar to that of *dystrophic* mice in terms of abnormal myelination. Sural nerve biopsies reveal hypomyelination or hypermyelination and reduction of large fibers, although naked axons are rare [38, 84–86]. It is not clear whether the spinal root in these patients have axon sorting defects. In most MD-CMD patients, the nerve conduction velocity is reduced [38, 86, 87]; therefore, the peripheral nerve abnormality most likely contributes to the phenotypes. These studies suggest that the peripheral nerve and skeletal muscle pathology could both be therapeutic targets for treating MD-CMD patients.

Conclusions

Recent studies using transgenesis and gene knockout technology have revealed important features of laminin functions in PNS development: (1) laminins can mediate axonal sorting and myelination without basal lamina formation, indicating that signals from laminins are important for Schwann cell development; (2) laminin-mediated axonal sorting enables Schwann cell access to axon-derived mitogens and triggers their proliferation; (3) laminins are required for the transition from premyelinating Schwann cell to promyelinating Schwann cell via the maintenance of Krox-20 expression and the downregulation of Oct-6; and (4) laminins play important roles in the long-term survival of Schwann cells. Further understanding of the precise signaling pathways of laminins in Schwann cell development, axon sorting, and myelination will facilitate the development of new efficient methods for treating peripheral neuropathies such as MD-CMD in humans.

Acknowledgements We thank Prabhjot Dhadialla and Dr. Erin Norris for the comments on the manuscript, and Dr. Sidney Strickland and Dr. Karen Carlson for the useful discussion. Work in our laboratory is supported by grants from the NIH (NS035704-08 and NS038472-07), the Adelson Program in Neuronal Repair and Rehabilitation, and the Muscular Dystrophy Association (MDA4066).

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