Two PDZ Domain Proteins Encoded by the Murine Periaxin Gene Are the Result of Alternative Intron Retention and Are Differentially Targeted in Schwann Cells*

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Periaxin was first described as a 147-kDa protein that was suggested to have a potential role in the initiation of myelin deposition in peripheral nerves based upon its abundance, cell type specificity, pattern of developmental expression, and localization (Gillespie, C. S., Sherman, D. L., Blair, G. E., and Brophy, P. J. (1994) Neuron 12, 497–508). Here we show that the murine periaxin gene spans 20.6 kilobases and encodes two mRNAs of 4.6 and 5.2 kilobases that encode two periaxin isoforms, L-periaxin and S-periaxin of 147 and 16 kDa respectively. The larger mRNA is produced by a retained intron mechanism that introduces a stop codon and results in a truncated protein with an intron-encoded C-terminus of 21 amino acids. Both proteins possess a PDZ domain at the N terminus; nevertheless, they are targeted differently in Schwann cells. Like other proteins that contain PDZ domains, L-periaxin is localized to the plasma membrane of myelinating Schwann cells; in contrast, S-periaxin is expressed diffusely in the cytoplasm. This suggests that proteins that contain this protein-binding module may also participate in protein–protein interactions at sites other than the cell cortex.

Periaxin was first identified as a relatively abundant 146-kDa protein of myelinating Schwann cells in a screen for novel cytoskeleton-associated proteins with a role in peripheral nerve myelination (1). Like P0, the major integral membrane protein of peripheral nervous system myelin, periaxin is detectable at an early stage of peripheral nervous system development (2). However, in contrast to P0, periaxin is not incorporated into compact myelin (1), but is initially concentrated in the plasma membrane, the abaxonal membrane (apposing the basal lamina), and the adaxonal membrane (apposing the axon). As myelin sheaths mature periaxin becomes concentrated in the abaxonal membrane and plasma membrane (2). This shift in the localization of the protein in the Schwann cell after completion of the spiralization phase of axon ensheathment suggests that periaxin may participate in the membrane-protein interactions that are required to stabilize the mature sheath. To shed light on the protein’s function, we were particularly interested to determine if modular protein-binding domains might be represented in the periaxin amino acid sequence. Although no such domains were identified from initial data base comparisons (1), here we report that the periaxin gene does encode one of the most interesting of these protein-binding motifs to emerge over recent years, namely the PDZ domain.

The PDZ domain was named after the three proteins in which it was first identified, namely post-synaptic density protein PSD-95, Drosophila discs large (dlg) tumor suppressor gene, and the tight junction-associated protein ZO-1 (3). It consists of an approximately 90-amino acid protein-binding motif found in proteins that interact with the cytoplasmic tail of plasma membrane proteins and with the cortical cytoskeleton (4). Although the binding site for some PDZ domains is the simple peptide sequence (S/T)XV found at the C terminus of certain plasma membrane proteins (5), PDZ-containing proteins can recognize somewhat different sequences and can even form homophilic clusters with the PDZ domains of other proteins (6, 7). So far, two major functions have been ascribed to PDZ domains on the basis of their interactions with plasma membrane proteins and their presence in known signaling molecules such as dlg (6). First, they may organize and recruit proteins to the plasma membrane as has been proposed for PSD-95 (3). Secondly, they may link transmembrane proteins with the actin cytoskeleton via actin-binding proteins such as protein 4.1 (8–10).

Here we report the structure of the murine periaxin gene, which reveals that there are two periaxin isoforms, L-periaxin and S-periaxin. The smaller protein, S-periaxin, is generated by a relatively rare retained intron mechanism (11), which is probably favored by the presence of suboptimal 5′- and 3′- splice sites in the final intron together with a downstream putative exonic splicing enhancer (12, 13). Although both are PDZ proteins, L- and S-periaxin are targeted differently in the Schwann cell, indicating that the PDZ domain is not the sole determinant of their subcellular localization. To our knowledge this is the first example of the differential localization of two protein isoforms with the same PDZ domain.

EXPERIMENTAL PROCEDURES

Isolation and Characterization of Genomic Clones—A 129SV mouse genomic library (gift from Dr W. Skarnes, Center for Genome Research, Edinburgh University) in the bacteriophage Lambda Dash II (Stratagene, Cambridge, UK) was screened by plaque hybridization (14) with a 32P-labeled probe comprising nucleotides 1–3000 of rat periaxin cDNA (1). Hybridization was at 60 °C in QuikHyb (Stratagene, Cambridge, UK), and the filters were washed in 2× SSC (300 mM NaCl, 30 mM sodium citrate, pH 7), 0.1% (w/v) SDS at room temperature and finally at 60 °C in 0.2× SSC, 0.1% SDS. From 1×106 clones, twelve positive plaques were identified after three rounds of screening, and EcoRI fragments of each insert were subcloned into the pHBl30 plasmid (IBI, Cambridge, UK) for analysis. The ends of each clone were sequenced by the dideoxy chain termination method (15) using a T7 DNA polymerase kit (Pharmacia LKB Biotech, Uppsala, Sweden), which showed that the twelve clones comprised three groups of sequences that overlapped (H1, F2, and I1). Oligonucleotide primers designed from these sequences were used to determine the order of the EcoRI fragments within the
gene by PCR using purified bacteriophage DNA (1 μl of 1:500 dilution) as template (first cycle of 94 °C for 5 min, 60 °C for 1 min, and then 72 °C for 1 min; 36 cycles of 94 °C for 1 min, 60 °C for 1 min, and then 72 °C for 1 min, 7 min during the last cycle). The reactions (50 μl) included primers (5 μM) and DyNazyme DNA polymerase (5 units) (Flankgen Ltd., Slough, UK). The PCR sequence was mapped to the EcoRI fragments by Southern blot with regional periaxin probes. Briefly, EcoRI-digested plasmid DNA from each EcoRI subclone was electrophoresed on a 1% agarose gel and vacuum blotted to Magna nylon membrane (Micron Separations Inc., Westborough, MA). The membrane was hybridized with 32P-labeled rat periaxin cDNA fragment covering the entire rat cDNA, and the order of the genomic clones was determined. The cDNA and genomic sequences were compared using the University of Wisconsin GCG software package. Analysis of the three clone types from the first screening revealed the absence of sequence corresponding to the first 297 bases of the rat cDNA, which includes the initiation codon and the 5′-untranslated region. Therefore a further 6 × 103 plaques were screened with a probe corresponding to nucleotides 50–297 of the rat sequence (1), which was generated by reverse transcription-PCR of the 5′-end of mouse periaxin mRNA (see below). Two clones were isolated and characterized as described above and shown to be identical. This sequence (HHI) did not overlap with the 5′-end of H1. However HH1 and H1 were shown to be contiguous by PCR performed on 128SV genomic DNA as template (first cycle of 94 °C for 5 min, 60 °C for 1 min, and then 72 °C for 1 min; 36 cycles of 94 °C for 1 min, 60 °C for 1 min, and then 72 °C for 1 min, 7 min during the last cycle). The reactions (50 μl) contained genomic DNA (200 ng) primers (5 μM) and DyNazyme DNA polymerase (5 units) (Flankgen Ltd.). The PCR product was isolated from an agarose gel with the QiAEX II gel extraction kit (Qiagen Ltd, Crawley, UK) cloned into the pGEM-T vector (Promega Ltd., Southampton, UK) according to the manufacturer’s instructions and sequenced using a T7 sequencing kit (Promega Ltd.), random hexamers (150 ng), dithiothreitol (10 mM), dNTPs (0.5 mM), and SuperScript reverse transcriptase (400 units) (Life Technologies Inc., Paisley, Scotland) in first strand synthesis buffer and were incubated at 42 °C for 1 h. The reactions were terminated by heating at 80 °C, and 1 μl of this reaction was used as a template for PCR using primers corresponding to nucleotides 50–71 and 651–650 of the published rat periaxin cDNA sequence (1). PCR conditions were five cycles of 94 °C for 1 min, 55 °C for 1 min, and 72 °C for 1 min (4 min during the last cycle). Reaction (50 μl) contained primers (5 μM) and DyNazyme DNA polymerase (1 unit) (Flankgen Ltd.). The product was subcloned into the pGEM-T-Vector (Promega Ltd., Southampton, UK) vector according to the manufacturer’s instructions and sequenced with a T7 DNA polymerase kit (Pharmacia).

Reverse Transcription-PCR—To obtain a probe for the 5′-end of the mouse periaxin mRNA, total RNA was isolated from the sciatic nerves of 15-day-old mice with RNeazol B (Biogenes, Bournemouth, UK) according to the manufacturer’s instructions. Reverse transcriptions (20 μl) contained RNA (2 μg), random hexamers (150 ng), dithiothreitol (10 mM), dNTPs (0.5 mM), and SuperScript reverse transcriptase (400 units) (Life Technologies Inc., Paisley, Scotland) in first strand synthesis buffer and were incubated at 42 °C for 1 h. The reactions were terminated by heating at 80 °C, and 1 μl of this reaction was used as a template for PCR using primers corresponding to nucleotides 50–71 and 651–650 of the published rat periaxin cDNA sequence (1). PCR conditions were five cycles of 94 °C for 1 min, 55 °C for 1 min, and 72 °C for 1 min (4 min during the last cycle). Reaction (50 μl) contained primers (5 μM) and DyNazyme DNA polymerase (1 unit) (Flankgen Ltd.). The product was subcloned into the pGEM-T-Vector (Promega Ltd., Southampton, UK) vector according to the manufacturer’s instructions and sequenced with a T7 DNA polymerase kit (Pharmacia).

Determination of Periaxin Transcriptional Initiation Site—An oligonucleotide complementary to nucleotides 50–71 of the rat periaxin cDNA (1) was end-labeled with [γ-32P]ATP and T4 polynucleotide kinase. Total RNA (10 μg) from the trigeminal nerves of 13-day-old mice was hybridized to labeled oligonucleotide (5 pmol) and reverse transcribed for 10 min at 80 °C followed by 18 h at 42 °C. Reverse transcription employed SuperScript reverse transcriptase (Life Technologies Inc.) in a buffer supplied by the manufacturer and included 50 μg/ml actinomycin D. Extended products were resolved on a 6% polyacrylamide DNA sequencing gel adjacent to a dyeoxy sequencing ladder comprising the 5′-end of periaxin primed using an oligonucleotide complementary to nucleotides 103–122 of mouse periaxin cDNA.

Isolation of cDNA Clones Encoding Two Periaxin Isoforms—Two periaxin mRNAs of 5.2 and 4.6 kb were detected in mouse and rat sciatic nerve. We had previously cloned the cDNA for the smaller periaxin mRNA (1). To identify how the larger mRNA differed from the 4.6-kb mRNA, 3 × 103 clones of a 15-day-old rat sciatic nerve cDNA library constructed in EcoRI-digested Agt11 (Promega Ltd., Southampton, UK) according to the manufacturer’s instructions were screened with a probe comprising nucleotides 1–597 of the rat periaxin cDNA (1) as described (1). Screening was carried out essentially as for the genomic library, and 24 positive plaques were purified. DNA from each of these phage clones was digested with NotI, and the inserts were subcloned into the pGEM11zf plasmid (Promega Ltd., Southampton, UK) and sequenced. Of 11 clones that included a sequence homologous to exon 6 of the mouse gene, five included a sequence corresponding to the intron between exons 6 and 7. Total RNA from both rat and mouse sciatic nerves was subsequently Northern blotted with a PCR-generated probe for this intron-sequence with identical results.

Northern Blotting—Total RNA from 15-day-old mouse sciatic nerves was electrophoresed on 0.8% agarose formaldehyde gels and transferred to Magna nylon membrane (Micron Separations Inc., Westborough, MA) by vacuum blotting in 20 × SSC. Filters were probed for 1.5 h in Rapid Hyb buffer (Amersham) at 65 °C with an exon 7-specific probe, a 1.3-kb restriction fragment corresponding to nucleotides 1–1324 of the rat periaxin cDNA sequence, or a PCR product homologous to the intron 6 of the mouse gene labeled with [α-32P]dCTP by random priming (Life Technologies Inc.) and were washed to a final stringency of 0.2 × SSC at 65 °C. The intron probe was prepared using one of the five rat cDNA clones isolated as described above as template with primers that flanked 400 bp of intron sequence. The PCR conditions used for the preparation of this intronic probe were: first cycle of 94 °C for 1 min, 55 °C for 1 min, and then 72 °C for 1 min; 26 cycles of 94 °C for 40 s, 55 °C for 1 min, and then 72 °C for 1 min, 4 min during the last cycle. Reaction (50 μl) contained primers (5 μM) and Dynazyme DNA polymerase (0.5 unit) (Flankgen Ltd.).

Antibody Production—The anti-SPeri and anti-NTerm antibodies were raised in rabbits against the synthetic peptides AKLVRVLSPYVPQDPSPDSRVAAC and EARSBSSEELRRRAC, respectively, which were generous gifts from Prof. N. Groome, Department of Biology, Oxford Brookes University. The former peptide corresponded to the C-terminal 23 amino acids of mouse S-periaxin, and the latter comprised the N-terminal 14 amino acids of mouse periaxin, which is identical in L-periaxin and S-periaxin. The C-terminal cysteine residue of each peptide was coupled to Keyhole Limpet hemocyanin by standard techniques and used to immunize rabbits. Anti-SPeri antibody was affinity purified by immobilization to a column of peptide coupled to Sepharose (Sigma Chemical Company, Poole, UK).

Immunofluorescence Microscopy—20-day-old mice were perfused intracardially with a 4% solution of paraformaldehyde in 0.1 M sodium phosphate, pH 7.4. Sciatic nerves were then removed and fixed for a further 2 h at room temperature. After washing, nerves were cryoprotected by immersion for 15 min in 5% (w/v) and then 10% (w/v) sucrose in 0.1 M phosphate, pH 7.4, followed by overnight incubation in a 20% (w/v) solution at 4 °C. Cryoprotected nerves were subsequently frozen in OCT embedding compound (Tissue TEK) using isopentane. Transverse sections (7 μm) were collected on 3-amino propyltriethoxysilane-subbed glass slides. Following removal of OCT by washing in phosphate-buffered saline (PBS) (Sigma), sections were blocked for 3 h at room temperature in a solution of 10% (w/v) goat serum (Scottish Antibody Production Unit, Law Hospital, Carluke, Scotland), 0.2% (w/v) gelatin, and 0.3% (w/v) Triton X-100 in PBS. Blocked sections were incubated overnight with rabbit anti-170pep1 (1) diluted 1:3000) or affinity-purified anti-SPeri (diluted 1:200) with mouse anti-myelin basic protein (diluted 1:200) from Prof. N. Groome, Department of Biology, Oxford Brookes University. Sections were then incubated in 1% rabbit anti-SPeri (diluted 1:20) and biotinylated goat anti-rabbit (Kirkegaard and Perry) (diluted 1:500). Both antibodies were in 4% (w/v) goat serum, 0.2% (w/v) gelatin, and 0.3% (w/v) Triton X-100 (in PBS) and incubated for 1 h at

**FIG. 1. Structure of the murine periaxin gene.** Exons are numbered and indicated by solid rectangles. Introns are shown as solid lines between the exons, except for intron 6, which is depicted as a hatched box between exons V and VI. EcoRI (E) restriction sites are indicated and were used to subclone the genomic clones F2, H1, and 11 (from the first screen) and HH1 (from the second screen).
The intronic 5’-splice donor GT and 3’-splice acceptor AG are in bold type. The exonic sequences are capitalized. The interruption of codons by introns is indicated by the phase. A phase of 0 indicates no interruption, and insertion of an intron after the first nucleotide of the codon is indicated by phase 1. All numbering is relative to the deduced cDNA sequence of the mouse 4.6-kb periaxin message.

| Exon no. (size in bp) | cDNA position | 5’-Splice donor | Intron no. (size in bp) | 3’-Splice acceptor | Codon phase | Amino acid at splice site |
|-----------------------|---------------|-----------------|-------------------------|------------------|-------------|-------------------------|
| 1 (32)                | 1–32          | CCACCGttaga     | 1 (4900)                | tctctagAGCCCCC   |             |                         |
| 2 (44)                | 33–76         | CCTACAGttaga    | 2 (88)                  | tgcgcgGACGCA     |             |                         |
| 3 (99)                | 77–175        | GACGCGtggg     | 3 (116)                 | ccccccGCTTAGG    |             |                         |
| 4 (126)               | 176–301       | GCTGAGtgggt    | 4 (2900)                | tgcgcgGAGCTG     | 0           | (Glu/Glu)*              |
| 5 (157)               | 302–458       | AAGAAAGtggcc   | 5 (7500)                | ttgtagGGGACC     | 1           | Gly*                   |
| 6 (197)               | 459–655       | AAGCTGtggc     | 6 (592)                 | cctctagAACATC    | 0           | (Leu/Asn)*              |
| 7 (4002)              | 656–4657      |                 |                         |                  |             |                         |

**Fig. 2.** Alternative splicing of a retained intron generates two mRNAs. Sciatic nerve RNA from 15-day-old mice was electrophoresed on a 0.8% agarose formaldehyde gel (2 μg/lane), transferred to nylon membrane and probed in lane A with a 1.3-kb restriction fragment of nucleotides 1–1324 of the rat periaxin cDNA and in lane B with a PCR product comprising intron 6 of the mouse gene. Both probes were labeled with [α-32P]dCTP by random priming as described under “Experimental Procedures.” Autoradiography was for 48 h.

**Fig. 3.** Determination of the periaxin transcription initiation site by primer extension. An oligonucleotide complementary to nucleotides 50–71 of rat periaxin cDNA (1) was end-labeled with [γ-32P]ATP and T4 polynucleotide kinase. Total RNA (10 μg) from the trigeminal nerves of 13-day-old mice was hybridized to labeled oligonucleotide (5 pmol) and reverse transcribed. Extended products were resolved on a 6% polyacrylamide sequencing gel adjacent to a sequencing ladder comprising the 5′-end of periaxin primed using an oligonucleotide complementary to nucleotides 103–122 of mouse periaxin cDNA. (lane P).

**Table I**

**Exon/intron position, size, and junction sequence structure of the mouse periaxin gene**

**RESULTS**

**Intron-Exon Structure of Murine Periaxin Gene**—Three different clones were isolated from a murine 129SV genomic library using a rat periaxin cDNA as a hybridization probe. A fourth, which included the 5′-end of the gene, was isolated using a murine cDNA probe that had been generated by reverse transcription-PCR. The clones were digested with EcoRI and subcloned into pBluescript. These clones were analyzed by PCR and Southern blot and were sequenced, which demonstrated that they encompassed the entire periaxin gene. The gene is divided into seven exons, and the coding region spans were approximately 20.6 kb (Fig. 1). Exons 1–6 range in size from 32 (exon 1) to 197 bp (exon 6). Exon 7 is the largest by far at 4002 bp. The
sizes of the introns were estimated by restriction mapping and PCR and range from 88 (intron 2) to 7500 bp (intron 5). The exon-intron boundaries were sequenced and the splice donor (GT) and acceptor (AG) sites were identified. This information is summarized in Table I.

Two distinct periaxin mRNAs are expressed in the rat peripheral nervous system (1), and the murine gene also encodes two mRNAs of 4.6 and 5.2 kb of approximately equal abundance (Fig. 2A). Of eleven cDNA clones isolated from a rat cDNA library, five included intron 6 (Fig. 1). Confirmation that the murine 5.2-kb mRNA differed from the 4.6-kb mRNA by the inclusion of this 592-bp intron was obtained by Northern blotting (Fig. 2).

Identification of Transcriptional Initiation Site and Core Promoter—The transcriptional initiation site was determined by primer extension and was found to be 75 bp upstream of a primer complementary to a region comprising the final nucleotide of exon 2 and the 5' 9-end of exon 3 in the murine gene (Fig. 3). This site lay in the sequence YYA1AGGA, which has some similarity to the sequence YYA1N(A/T)YY believed to be the consensus transcriptional initiator for RNA polymerase II transcripts (16). An identical transcriptional initiation site was found for the rat mRNA (data not shown). Approximately 500 bp of the putative core promoter was sequenced (Fig. 4). Though it lacks a TATA box, the promoter does possess a CAAT box, and between this motif and the initiator the sequence is relatively GC-rich (68%), which is commonly the case in TATA-less promoters (17). A sequence motif corresponding to the SCIP/Oct-6 binding site (position −241) is of particular interest owing to the role that Oct-6 is believed to play in Schwann cell maturation (18–20). An element (GCRE) at position −360 has previously been identified as mediating the induction of several myelin protein genes by forskolin (21). The presence of this sequence would help to explain the ability of cAMP to mimic some of the axonal signals that regulate the expression of differentiation-specific genes in Schwann cells (22, 23).

Deduced Amino Acid Sequence of L-Periaxin and S-Periaxin—the deduced amino acid sequence of the protein encoded by the 4.6-kb mRNA is depicted in Fig. 5A. This isoform, termed L-periaxin, is 93% identical to rat periaxin and has a size of 147.500 kDa, slightly larger than the rat protein (1). The presence of a retained intron in the larger 5.2-kb mRNA introduces a stop codon preceded by a sequence that encodes a unique 21-amino acid C terminus. This truncated isoform, termed S-periaxin, has a size of 16.2 kDa (Fig. 5B). Except for two differences at the extreme C terminus, the rat and murine S-periaxins are identical. Anti-peptide antibodies recognizing the N terminus of L- and S-periaxin (anti-NTerm), the repeat region unique to L-periaxin (anti-170pep1 (1)), or the C terminus unique to S-periaxin (anti-SPeri) confirmed the structural relationship between the two isoforms (Fig. 6).
examine this sequence very carefully for conserved motifs that might illuminate the function of these proteins, despite the fact that previous searches of the complete L-periaxin polypeptide had not been informative (1). A degree of sequence similarity with a portion of the PDZ domain of the junction-associated protein ZO-1 provided the necessary clue. Comparison of the N terminus sequence from amino acids 13 to 97 with several well characterized PDZ domains in other proteins confirmed that this region comprises a PDZ domain common to both L- and S-periaxin (Fig. 7).

Because proteins that contain PDZ domains are believed to associate with the plasma membrane (3, 6), it was of considerable interest to compare the subcellular locations of L- and S-periaxin in the Schwann cell. In transverse sections of sciatic nerves from 20-day-old mice L-periaxin was detected in a typical annular pattern that reflected its concentration in the periaxonal and abaxonal myelin lamellae (Fig. 8, A and C). In contrast to myelin basic protein, L-periaxin was also present at the plasma membrane of the Schwann cell, consistent with the possession of a PDZ domain. As found before, L-periaxin is not present in compact myelin where myelin basic protein is abundant (Fig. 8, B and C) (1, 2). In contrast, S-periaxin was not concentrated at the interface between the plasma membrane and cytoplasm of the Schwann cell or the myelin sheath (Fig. 8, D–F). Instead, this protein seemed to be distributed fairly evenly throughout the cytoplasm. There also appeared to be some S-periaxin in the nucleus of the Schwann cell. Apparently, the presence of a PDZ domain may not be sufficient to direct the association of S-periaxin with either the Schwann cell plasma membrane or its product, the myelin sheath.

**DISCUSSION**

One of the most interesting aspects of the expression of the periaxin gene is the fact that alternative splicing involves the retention of an intron. There are approximately equal proportions of two mRNAs that either lack or include the last intron. The factors that determine the retention of introns have been studied in detail by Rottman and colleagues, and a key feature is the “weakness” of the splice sites at the 5’- and 3’-ends of the intron, i.e. the extent to which they differ from the consensus sequences that are known to promote splicing (12, 24). These weak splice sites would not normally support splicing without an additional sequence in the downstream exon called the exonic splicing enhancer (13). In support of this model, the splice sites in the last intron of the periaxin gene are divergent from the consensus sequences, and there is a domain downstream in exon 7 that is highly purine-rich. The sequences of these 5’- and 3’-splice sites are CTGgtagtcg and tcagA, respectively (intron sequences in lowercase), which are significantly different from the corresponding consensus donor and acceptor sequences of CAGgtagtcg and ncaagG (24). Downstream of the acceptor site and 24 bases into exon 7 there is a 14-base sequence GAAGAAGAA-GAAGA, which is an excellent candidate for an exonic splicing enhancer (1, 13).

Of those genes that are typically expressed by myelin-forming Schwann cells, the periaxin gene is one of the first to

![FIG. 6. Western blot of mouse periaxin with domain-specific antibodies.](image1)

Mouse sciatic nerves from 15-day-old mice were homogenized and immunoblotted using antibodies raised against peptide sequences comprising either the N terminus of L- and S-periaxin (anti-NTerm), the repeat region unique to L-periaxin (anti-170pep1 (1)) or the C terminus unique to S-periaxin (anti-SPeri). The sizes of L- and S-periaxin are indicated in kDa.

![FIG. 7. Identification of a PDZ domain at the N terminus of L- and S-periaxin.](image2)

The sequence of mouse L- and S-periaxin between amino acids 14 and 98 was compared with PDZ domains in the Caenorhabditis elegans protein CET19B10 (residues 232–316) (32), PSD-95 (residues 309–393) (33), Discs-Large (dlg) (residues 482–566) (34), SAP97 (residues 461–545) (35), ZO-1 (residues 408–491) (36), ZO-2 (residues 93–176) (37), and inaD (residues 361–447) (38). The eight segments within the domain comprising six β strands and two α-helices are as determined and described by Doyle et al. (5). The sequences have been arranged to maximize their similarity. Solid blocks show amino acid identity, and shading indicates conservative substitutions.
PDZ Domains and Periaxin Targeting

Vital to maintain periaxin expression in Schwann cells the axon (21). Certainly axonal contact has been shown to be cAMP can mimic to some extent the inductive interactions of ing site found in other genes that are characteristically ex-

The discovery of a PDZ domain in the periaxin proteins is certainly to provide exciting new ways of interpreting the function not only of periaxin but also of PDZ domain-containing proteins in general. The PDZ motif in periaxin may either participate in the membrane-protein interactions that are required to promote spiralization or act to recruit proteins to a cortical scaffold important in transmembrane signaling, at least for L-periaxin. The fact that S-periaxin is not concentr-

In conclusion, we have shown that two periaxin isoforms, both of which contain the same PDZ domain, are generated in approximately equal amounts by an alternative retained intron mechanism. Furthermore, these PDZ proteins are uniquely targeted to different localizations in myelinating Schwann cells, which suggests that the presence of a PDZ motif may not be the dominant determinant in the selection of binding partners for L- and S-periaxin in myelinating Schwann cells.

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