Genomic Organization of the Human LAR Protein Tyrosine Phosphatase Gene and Alternative Splicing in the Extracellular Fibronectin Type-III Domains*

(Received for publication, May 31, 1994, and in revised form, July 18, 1994)

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The structure of the human leukocyte-common antigen-related molecule (LAR) protein tyrosine phosphatase gene was elucidated using phage and cosmide genomic DNA clones. The LAR gene is composed of 33 exons spanning over 85 kilobase pairs. Exon 2 encodes the signal sequence and the first four amino acids in the mature LAR protein. The three immunoglobulin-like domains are encoded by exons 3-7, and the eight fibronectin type II (Fn-III) domains by exons 8-17. Exons 18-22 encode the juxtamembrane and transmembrane domains, and exons 23-33 encode the two conserved tyrosine phosphatase domains and the entire 3' untranslated region. Exon 1, which presumably encodes the 5' untranslated sequence, has not been identified. Reverse transcription-polymerase chain reaction analysis revealed the alternative splicing of a mini-exon (exon 13) in the Fn-III domain 5 of human LAR and other related genes (rat LAR, rat PTPα, and human PTPβ). RNase protection analysis showed that the human LAR mRNA in which exon 13 is spliced-out is the major mRNA species in all cell lines examined. Reverse transcription-polymerase chain reaction analysis revealed further alternate splicing of LAR mRNA involving the Fn-III domains 4, 5, 6, and 7 in various combinations. These findings will facilitate the understanding of the physiological functions of the LAR extracellular domain.

The control of protein tyrosine phosphorylation is vital in the regulation of normal physiological processes and its deregulation is implicated in the oncogenic process. Reversible tyrosine phosphorylation is achieved through the interplay of both protein tyrosine kinases and protein tyrosine phosphatases (PTPases). The members of the PTPase family, which is distinct from the protein serine/threonine phosphatase families, include both cytosolic and transmembrane forms (1-3). PTPases all have in common a highly conserved catalytic domain. Cytosolic PTPases have a single PTPase domain, whereas receptor-like, transmembrane PTPases may have one, but more commonly have two, tandem PTPase domains. The extracellular regions of the transmembrane PTPases are very diverse and can be divided into a number of subfamilies. These extracellular regions can be further diversified by alternative splicing. For example, at least five different isoforms of CD45 that are generated by alternative splicing are differentially expressed during development and according to cell type (4). Furthermore, isolation of variant cDNA clones suggests alternative splicing in the extracellular domain of the transmembrane PTPases PTPβ, PTPα, PTPγ, and PTPρ (PTPρ is also known as PTP-11/FS, PTP-NE3, and rat LAR PTP-2) (5-11).

LAR (leukocyte-common antigen-related molecule) is a prototype for a family of transmembrane PTPases whose extracellular regions are composed of a combination of immunoglobulin (Ig)-like domains and fibronectin type III (Fn-III) domains (12, 13). The close similarity of the extracellular region of LAR with the neural cell-adhesion molecule (NCAM) raised the prospect that LAR may also function in cell adhesion and that this protein could thus directly link extracellular adhesion with intracellular signal transduction. LAR cDNA clones have been isolated from human and rat, and their deduced amino acid sequences are 96% identical (12, 14). Furthermore, two other mammalian transmembrane PTPases (PTPα and PTPρ) and a Drosophila PTPase (DLAR) are closely related to LAR (5, 8-10, 15). cDNA isoforms of LAR-related proteins have been reported suggesting alternative splicing of the extracellular domain in the Fn-III and the Ig-like region as well as in the PTPase domains (5-11).

The LAR protein has a broad tissue distribution and is expressed on cells of many different lineages including epithelial cells, smooth muscle cells and cardiac myocytes (13). LAR is synthesized as a >200-kDa precursor which is cleaved by an endogenous protease into two subunits (150 and 85 kDa) that remain non-covalently attached. The 150-kDa extracellular subunit is subsequently shed during cell growth (13). Clearly LAR is a complicated protein composed of a number of different protein modules. To understand the evolutionary relationship of these modules to other related molecules, knowledge of the genomic organization is necessary. Such knowledge will also facilitate the structure-function study of the LAR PTPase. To date, however, the gene structure is known for only two transmembrane PTPases, i.e. CD45 and PTPα, both of which lack extracellular Ig-like and Fn-III domains (16-19). In this report, we describe the exon/intron organization of the human LAR gene, which is located on chromosome 1, region p32-33 (13). We also report the alternative usage of several exons that encode the extracellular Fn-III domains.

MATERIALS AND METHODS

Screening of Genomic DNA Libraries—Human genomic DNA libraries in the λ phage vectors, Charon26, EMBL3, and EMBL3-SP6/77 were obtained from, respectively, P. Leder (20), S. Orkin (21), and Clontech

*This work was supported in part by National Institutes of Health Grants AI-29398 and CA-51132, and DFCI/Sandoz Drug Discovery Program Grant 91012. The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked "advertisement" in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

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The abbreviations used are: PTPase, protein tyrosine phosphatase; LAR, leukocyte-common antigen-related molecule; Fn-III, fibronectin type III; N-CAM, neural cell-adhesion molecule; RT, reverse transcription; PCR, polymerase chain reaction; Pipes, 1,4-piperazinediethanesulfonic acid; bp, base pair(s); kb, kilobase pairs.
**Fig. 1. Organization of the human LAR gene.** The exon/intron organization of the human LAR gene is schematically shown. The positions and relative sizes of the exons are indicated by boxes and the exons are numbered relative to the putative translation initiation site. Introns are indicated by horizontal bars. A restriction map of the LAR gene is shown, and recognition sites for the restriction enzymes EcoRI and HindIII are indicated by vertical bars. The cosmid and phage clones used for mapping and sequencing the exons are shown underneath the restriction map.

(catalog no. HL10765). These libraries were screened by filter hybridization using α-32P-labeled nick-translated LAR cDNA probes (12). Approximately 2 x 10^6 plaques were screened with each probe. Hybridization was carried out as described previously (5) except that the final washes were done in 0.5 x SSC and 0.1% SDS at 60 °C. A human placenta genomic library in the cosmID pWE15 vector (Stratagene) was also screened using LAR cDNA probes. Unique phage and cosm id clones were identified by restriction mapping and analyzed by Southern hybridization of restriction fragments with the appropriate LAR cDNA probes. Positively hybridizing restriction fragments were subcloned into appropriate plasmid vectors for further restriction mapping, Southern analysis, and DNA sequencing. DNA sequences were determined by the dyelex chain termination method using modified T7 DNA polymerase (22, 23). A human placenta genomic library in the λ EMBL3 SP6/T7 vector (Clonetech) was screened with oligonucleotide probes whose sequences were derived from the 5'-untranslated region of the LAR cDNA (positions 41-56 and 277-295). However, no specific hybridization was detected.

**Cell Culture**—The following cell lines were obtained from ATCC: HeLa (human cervical epithelioid carcinoma), U-373 MG (human glioblastoma), Hep G2 (human hepatocellular carcinoma), T-47D (human breast carcinoma), A-431 (human epidermoid carcinoma), CRL1885 (human skin fibrosarcoma), MIA Paca-2 (human pancreatic carcinoma), SW480 (human colorectal carcinoma), HaCaT (human keratinocyte), REX 10 (human PTP1A(-)), 1028-1047, 5'-AAGGCCACGTCGGAGTCTACATGC); oligo 9 (human LAR exon 12(+), 2510-2529 + a 5' EcoRI site, 5'-GGGTAATGCTGATGCTGCTGCTG); oligo 10 (human LAR exon 13(+), 2562-2679, 5'-AGAGGAGTCCGAGGCTAG); oligo 5 (human LAR exon 13(-)), 2562-2679, 5'-TCATTCCTGGAGCTCCCTCGTC); oligo 6 (human LAR exon 14(-)), 2709-2728 + a 5' XbaI site, 5'-CGCATCAAGGACATCATGCT); oligo 11 (rat PTP1A(+), 2279-2299, 5'-AGCCAATGTTTCGAGTCTG); oligo 12 (rat PTP1A(-), 2521-2540, 5'-TCATTCCTGGAGTCCGAGGCTAG); oligo 9 (human PTP1A(+) exon 1(+) exon 4(+), 2590-2610, 5'-CTTCTGACGATGCTG); oligo 7 (human LAR exon 1(+) exon 4(+), 2590-2610, 5'-TCATTCCTGGAGTCCGAGGCTAG); oligo 8 (human LAR exon 1(+) exon 4(+), 2590-2610, 5'-TCATTCCTGGAGTCCGAGGCTAG); oligo 9 (human PTP1A(+) exon 1(+) exon 4(+), 2590-2610, 5'-TCATTCCTGGAGTCCGAGGCTAG); oligo 10 (human PTP1A(-) exon 1(-) exon 4(-), 2590-2610, 5'-TCATTCCTGGAGTCCGAGGCTAG); oligo 11 (rat PTP1A(+) exon 1(+) exon 4(+) exon 8(+) exon 9(+) exon 14(+), 2590-2610, 5'-TCATTCCTGGAGTCCGAGGCTAG); oligo 12 (rat PTP1A(-) exon 1(-) exon 4(-) exon 8(-) exon 9(-) exon 14(-), 2590-2610, 5'-TCATTCCTGGAGTCCGAGGCTAG); oligo 13 (rat PTP1A(+) exon 1(+) exon 4(+) exon 8(+) exon 9(+) exon 14(+), 2590-2610, 5'-TCATTCCTGGAGTCCGAGGCTAG); oligo 14 (rat PTP1A(-) exon 1(-) exon 4(-) exon 8(-) exon 9(-) exon 14(-), 2590-2610, 5'-TCATTCCTGGAGTCCGAGGCTAG).

**RESULTS**

**Isolation and Characterization of LAR Genomic Clones**—Screening of human placental genomic libraries, made in λ phage vectors, with LAR cDNA probes yielded 14 positive clones, determined to be unique by restriction mapping. Nine of these clones were further characterized as shown in Fig. 1. The two most 5' clones overlapped with each other and the six 3' clones also overlapped with each other. One clone, LAR-G28, did not overlap with any other clone. These nine λ clones together contained most of the LAR coding sequence. Screening of a human placental genomic library yielded six unique cosm id clones. Two of these encompassed all of the missing exons except for the 5' untrans lated region of the cDNA (Fig. 1). The numbering of the exons in Fig. 1 is based on the assumption that only one exon encodes the 5'-untranslated region.

**Structure of the Human LAR Gene**—Phage and cosm id clones that hybridized with LAR cDNA probes were further characterized by detailed restriction mapping, Southern analysis and DNA sequencing. The organization of the LAR gene is shown in Fig. 1 and the exon size and exon/intron junctional sequences are detailed in Fig. 2. The LAR gene spans at least 85 kb and is composed of 33 exons. A large intron of at least 16 kb separates exon 2 from the putative exon 1. Exon 2 encodes the signal peptide and the first 4 amino acids of the mature LAR protein. Exons 3-7 code for the three Ig-like domains of LAR, exons 9-17 for the eight Fn-III domains, exons 18-20 for the juxtamembrane and transmembrane regions, and exons 21-33 for one 2TPase domain. Exon 33 also encodes the entire 3'-untranslated region.

There is considerable variation in the arrangement of the exons within the gene. The exons encoding the Ig-like domains span the largest region of the gene with introns at least as big as 10 kb. In contrast, the exons encoding the two 2TPase domains are clustered together with 11 exons spanning 6.5 kb. All of the exon/intron junctional sequences are in agreement with the consensus donor and acceptor splice sequences (Fig. 2).

We observed several polymorphic differences in the nucleotide sequence. Cytosine at position 2275 of the LAR cDNA (nucleotide positions 2557-2908) in the reverse orientation between the HincII and PstI sites in the pSP65 vector (26). After linearization of pLAR-RP with PstI, a uniformly labeled antisense RNA probe was synthesized from the SP6 promoter with 20 units of SP6 RNA polymerase and [α-32P]CTP (800 Ci/mmol) as described previously (26). This probe (6 x 10^6 cpm) was hybridized with 50 μg of total cellular RNA overnight at 45 °C in hybridization buffer (40 mM PIPES, pH 6.4, 400 mM NaCl, 1 mM EDTA, 80% formamide). RNase digestion was carried out with 40 μg/ml RNase A and 230 units/ml RNase T1 at 30 °C for 1 h, followed by treatment with 20 μg of proteinase K in 0.5% SDS for 30 min at 37 °C, phenol extraction, and ethanol precipitation. Samples were analyzed on a 6% acrylamide urea sequencing gel. Molecular weight standards were made by end-labeling a 123-bp DNA ladder (Life Technologies, Inc.) with [α-32P]CTP and the Klenow enzyme.
C$^{017}$, C$^{116}$, and A$^{253}$ of the cDNA sequence were G, A, and G, respectively, in the genomic DNA clones. There is also an insertion in the 3' untranslated region of the genomic DNA where a T in the cDNA is replaced by a 23-nucleotide sequence, CTGAGTTCACTTTGGATCCTA, in the genomic DNA.

Analysis of the Exon Structure of the LAR Protein Domains—
The relationship of the exon structure of LAR with its protein domains is presented in Fig. 3A. The LAR protein contains three Ig-like domains at the N terminus. The first two of these are each composed of two exons, each exon containing one of the conserved cysteine residues. The entire third Ig-like domain is encoded by one exon. Large introns separate each of the Ig-like domains. However, within the domains the intron sizes vary greatly: the intron between exons 3 and 4 is only 163 bp, but the intron between exons 5 and 6 is ~10 kb. The last Ig-like exon is within 2 kb of the first Fn-III domain exon. The ten exons encoding the eight Fn-III domains of LAR extend over 15 kb of genomic DNA (Fig. 1). There is a good correlation between exon structure and the boundaries of Fn-III domains, although there is variation between the number of exons per domain. Fn-III domain 3 is encoded by 2 exons whereas only one exon encodes Fn-III domain 4. Two exons encode, respectively, two Fn-III domains: exons 6 and 7. The genomic structure of Fn-III domain 5 is unusual because this domain is encoded by three exons, one of which (exon 13) is only 27 bp in length. The eighth exon, which also includes non-Fn-III sequences, is only 27 bp in length. The eighth exon, which also includes non-Fn-III sequences, is only 27 bp in length. The eighth exon, which also includes non-Fn-III sequences, is only 27 bp in length. The eighth exon, which also includes non-Fn-III sequences, is only 27 bp in length.
FIG. 3. **Relationship of the exon structure and protein domains.** A, the exon organization of the LAR protein Ig-like domains, Fn-III domains, transmembrane region, and tandem PTPase domains are shown. In each case the exon/intron structure is indicated on top with the corresponding LAR protein domains shown schematically below. B, the exon organization of the tandem PTPase domains of the LAR gene is compared to those of the CD45 and PTPα genes (16-19). Each box represents an exon and the horizontal bar an intron (not drawn to scale). The shading in the most 3′ exons indicates the 3′-untranslated regions.

![Diagram](image)

FIG. 4. **PCR analysis of alternative splicing of exon 13 in the Fn-III domain 5 of LAR and LAR-related proteins.** A, total RNA isolated from the indicated human cell lines was reverse transcribed and PCR amplified using oligonucleotide primers 2 and 6, which correspond to sequences in the Fn-III domain 5, and flank exon 13 (see “Materials and Methods”). B, total RNA from rat and human cells was RT-PCR-amplified with the oligonucleotide primers 11 and 12 (for rat LAR), 13 and 14 (for rat PTPo), or 9 and 10 (for human PTPβ), which are derived from sequences in the Fn-III domain 5 and flank the putative exon 13 sequence. In both A and B, schematic outlines of the exon structures of the bands, determined by the nucleotide sequencing of subclones, are shown at the right, and the positions of markers are indicated at left. C, nucleotide (nt) and deduced amino acid sequences of human LAR, rat LAR, human PTPβ, and rat PTPo, with and without the exon 13 sequence. Sequences around the splice junctions are shown. Sequences that contain the exon 13 sequence are identical to the previously reported sequences (5, 11, 12, 14).
conservation of exon/intron junctions 28/27 and 31/32 in the LAR gene and the homologous junctions in the CD45 and PTP\alpha genes. These junctions interrupt the highly conserved sequence VHC\textsubscript{SA}G\textsubscript{V} (HICRD\textsubscript{OS} in D2 of CD45) that includes the catalytic center of the enzyme (28–30). This sequence, in each PTP\alpha domain, is exactly split by an intron in phase 2 in the fourth amino acid.

**Alternative Splicing of Exon 13**—Exon 13, which encodes a part of Fn-III domain 5, is unusually small compared to the other LAR exons (see Fig. 2). This exon codes for 9 amino acids that interrupt the alignment of Fn-III domain 5 with other Fn-III domains and make this domain longer than the other Fn-III domains. This sequence is missing in the reported rat LAR sequence, which is otherwise 96% identical with human LAR, suggesting that exon 13 is either specific for human LAR or that it is alternatively spliced (14). Excision of this exon would not interrupt the reading frame of the protein. Thus, we examined whether exon 13 in human LAR is alternatively spliced. Oligonucleotide sequences derived from exons 12 and 14 were used as primers for RT-PCR of RNA isolated from a number of cultured human cell lines (Fig. 4A). Two PCR products were observed in all cell lines examined. Cloning and DNA sequencing of the PCR products confirmed that the smaller product (196 bp) indeed represented LAR cDNA in which exon 13 was exactly spliced-out (Fig. 4C). The larger product (223 bp) was the form of LAR containing the exon 13 sequence. To determine if this alternatively spliced exon 13 is specific for human LAR or is a general phenomenon for all LAR-related proteins, the homologous region of rat LAR, human PTP\beta, and rat PTP\alpha was analyzed for the presence of an alternatively spliced exon. To this end, oligonucleotide sequences in the regions flanking the putative splice site of these proteins were used as primers for RT-PCR. Sequencing of the PCR products shown in Fig. 4B confirmed that the exon 13 sequence can be either present (Fig. 4B, upper band) or spliced-out (Fig. 4B, lower band) for all of these genes (Fig. 4C). The amino acid sequence of the insert in rat LAR is identical to human LAR, although some silent nucleotide differences are observed. The sequence of the insert in rat PTP\alpha is more similar to that of human PTP\beta than human LAR (Fig. 4C). All four sequences are negatively charged with the positions of the charged residues being conserved among the proteins.

**RNase Protection Analysis of Alternative Splicing of LAR**—To assess more quantitatively the abundance of the LAR mRNA isoform that contains the exon 13 sequence, an RNase protection analysis was performed. The antisense RNA probe spans exons 12 through 15 (Fig. 5, bottom). This probe was used in an RNase protection assay using total RNA derived from a wide range of human tissue cultured cells. As can be seen in Fig. 5, top, the strongest protected band is 228 nucleotides which represents LAR mRNA in which exon 13 is spliced-out. The band is a doublet probably due to heterogeneity of RNase digestion. The LAR mRNA isoform containing the exon 13 sequence is represented by the band at 352 nucleotides and is clearly the minor form. Longer exposure of the autoradiograms revealed that this band was present in most of the cell lines examined. Although no bands were visible for T and B cell lines (REX and CESS, respectively) in Fig. 5, the use of a higher amount of RNA (150 \mu g), and longer exposure times, did reveal a protected band of 228 nucleotides (data not shown).

**Multiple Alternative Splicing in the Fn-III Domains of LAR**—An alternatively spliced mini-exon within a Fn-III domain is a unique finding, not just for LAR but for any Fn-III containing protein. However, alternative splicing of an entire Fn-III domain, or domains, has been reported for other proteins such as fibronectin and tenasin (31, 32). Also, cDNA isoforms of mouse PTP\beta and rat PTP\alpha have been found that lack some of the Fn-III domains, suggesting a process of alternative splicing (6, 8–10). Accordingly RT-PCR was performed on a number of human cell lines using probes derived from the LAR gene exons 10, 12, 13, 14, 15, and 16. This analysis demonstrates that exon 14 can be spliced either to exon 15, or directly to exon 16 (Fig. 6). Splicing of exon 14 to exon 16 results in the precise deletion of Fn-III domains 6 and 7, but otherwise the proper reading frame is maintained. A LAR mRNA isoform containing exons 14, 15, and 16 could not be detected by the primers used in Fig. 6C, presumably because the size of the product is too large for amplification (data not shown). However, PCR primers derived from exons 15 and 16 did detect the joining of exon 15 to exon 16 (data not shown). It was also found that exon 10 can be directly spliced to exon 12 (Fig. 6, D and E, upper bands). This splicing event results in the precise elimination of Fn-III domain 4 without interrupting the reading frame. Finally, two further alternatively spliced mRNA isoforms, albeit at quite low abundancies, were observed in this region of LAR in which exon 10 is spliced directly either to exon 13 or to exon 14 (Fig. 6, D and E, respectively, bottom band). These splicing events would affect the reading frame and result in truncated protein products. This might be reminiscent of the recently reported case of the PTP\gamma gene. PTP\gamma is a transmembrane PTP\alpha in which an alternative splicing event in the extracellular region...
FIG. 6. Multiple alternative splicing of the LAR extracellular domain. RT-PCR was performed on total RNA isolated from the indicated cell lines. Oligonucleotides used as primers are: A, 3 and 7; B, 4 and 7; C, 3 and 8; D, 1 and 5; and E, 1 and 6 (see "Materials and Methods"). PCR products were analyzed on a 4% NuSieve-agarose gel and detected by ethidium bromide staining and UV light. The position and orientation of the sense and antisense oligonucleotide primers used, in relation to the LAR exon structures, are shown at right. Also shown at right are the schematic structures of the PCR products as determined by subcloning and sequencing of selected bands.

produces a truncated product that is a secreted extracellular matrix proteoglycan called phosphacan (33).

DISCUSSION

The exon/intron organization of the human LAR gene elucidated in this study reveals some interesting evolutionary aspects of this protein. The majority of genes encoding Ig-like domains have one exon per Ig-like domain, while a few, such as N-CAM, have two exons per Ig-like domain (34). The LAR gene has characteristics of both groups with the first two Ig-like domains encoded by two exons each, and the entire third Ig-like domain encoded by only one exon. This mixture of exon types has been reported for only a few other genes such as perlecan and DCC (35, 36).

Although Fn-III domains have been found in a wide range of proteins, very few have been analyzed at the genomic level. However, a general rule seems to be that a mammalian Fn-III domain is encoded by either 1 or 2 exons (37). In all known cases, including LAR, the exon/intron junction corresponding to a Fn-III domain boundary is phase 1. When two exons encode a Fn-III domain, an intron interrupts the coding region in the central, relatively nonconserved part of the domain, and the exon/intron junction may be in any phase. Within a gene that encodes multiple Fn-III domains, exon organization may be all of one type, e.g. N-CAM (2 exons per Fn-III domain), or a mixture of both types, e.g. tenascin (32, 38). Although LAR has a high sequence similarity with N-CAM, only Fn-III domains 3 and 8 are encoded by two exons each. The LAR gene is the first reported mammalian gene in which more than one fibronectin domain is encoded by a single exon. Two such exons are found in LAR: exon 8 encodes Fn-III domains 1 and 2, and exon 15 encodes Fn-III domains 6 and 7. There is only one LAR Fn-III domain (domain 4) that is encoded by a single exon. LAR exon 18 encodes both Fn-III and some non-fibronectin sequences. The fifth Fn-III domain of the LAR gene is highly unusual in that it is encoded by three exons (discussed in detail below).

Although the extracellular regions of transmembrane PTPases are dissimilar, the intracellular tandem PTPase domains are quite closely related. The exon/intron structure of the CD45 gene showed that PTPase domains 1 and 2 have a very similar exon/intron organization and probably arose by duplication (16-18). In general, the exon/intron structure of the PTPase domains are very similar among LAR, CD45, and PTPα. It appears, however, that either exon fusion has occurred in the LAR gene or intron gain has occurred in the CD45 and PTPα genes. This may be reflected in the fact that the genomic DNA encoding the two LAR PTPase domains spans only 6.5 kb, whereas the homologous region of CD45 and PTPα is >20 kb. The regions of greatest exon diversity are in the N-terminal region of both PTPase domains, and they may be regions determining specificity of the different phosphatases. A unique acidic insert is seen here in the second PTPase domain of CD45, and a unique alternatively spliced exon is seen in this region in the first PTPase domain of PTPα. The catalytic site of the PTPase domain, which has the consensus sequence VHCSAGV, is interrupted by an intron at exactly the same position in the three transmembrane PTPases. However, an exon/intron junc-
tion is not observed at this position in the cytoplasmic PTPase PTP1B (39). This observation points to an early evolutionary divergence of the cytoplasmic and transmembrane PTPases. A comparative analysis of the PTPase amino acid sequences has led to a similar conclusion (5).

A small exon was found in Fn-III domain 5, which is alternatively spliced in human LAR and other related proteins (rat LAR, rat PTPor, and human PTP6). Conservation of the exon 13 alternative splicing at the homologous position in these genes suggests an important physiological role for these nine amino acid sequences that share similar chemical characteristics. A comparison of the LAR Fn-III domain 5 sequence with the crystallographic structure of a Fn-III domain indicates that the exon 13 encoded nine-amino acid peptide is in a relatively non-conserved loop region (40). This, and the fact that this peptide is highly charged, suggest that it is at the surface of the Fn-III domain and thus that it is possibly involved in ligand interaction.

Where alternative splicing of Fn-III domains has been reported for other proteins such as fibronectin and tenascin, one small exon was found in Fn-I11 domain with its extracellular domain with its gene, LAR, (1994) Semin. Cell Biol. 5, 379-387. At this position in the cytoplasmic PTPase domain 4-7. Although our RT-PCR analysis of Fn-I11 domains 4-7. Although our RT-PCR analysis of Fn-I11 domains 4-7.

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