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JOURNAL ARTICLE

Mouse testicular sulfated glycoprotein-1: sequence analysis of the common backbone structure of prosaposins

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We have generated a cDNA encoding the mouse sulfated glycoprotein-1 (SGP-1) by polymerase chain reaction amplification of a mouse testicular Uni-Zap XR cDNA library with two synthetic oligonucleotide primers. A positive signal of 1,959 bases was isolated and subcloned into the pGEM-T. Sequence analysis showed a near identical nucleotide and amino acid similarity to mouse prosaposin cDNA. A few amino acid differences were found, and they may represent strain-specific heterogeneities. The cDNA has 88% amino acid identity to rat SGP-1 and 64% identity to human prosaposin. Prosaposin is the precursor of four lysosomal saposins (A, B, C, and D) that are generated by the proteolytic maturation of the former. Saposins are sphingolipid binding proteins that function as activators of lysosomal enzymes involved in sphingolipid hydrolysis. Northern blot analysis demonstrated that SGP-1 mRNA is transcribed in the seminiferous epithelium by Sertoli cells but not by germinal cells. Our results also demonstrated two forms of alternatively spliced testicular SGP-1 mRNA. This alternative splicing results in the inclusion or exclusion of exon 8, which encodes for three amino acid residues (QDQ) that are implicated in the sphingolipid binding affinity of saposin B. Sequence aligment indicates that all saposins share a common motif characterized by six conserved cysteines, a conserved N-linked glycosylation site, a conserved proline residue, and 15 positions that are characterized by large hydrophobic amino acids. These characteristics, together with similar secondary structure predictions and the predicted similar formation of three disulfide linkages, create a common framework of amino acids of three alpha helices enclosing an internal hydrophobic core for all saposins. The disulfide placement data, the hydropathy profile, and the presence of amphiphatic helices indicate that all saposins are stable proteins sharing similar secondary and tertiary structures.

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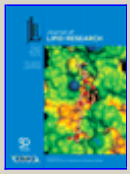

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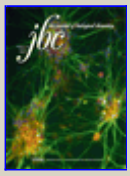

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