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Molecular Biology of Cardiac Sarcoplasmic Reticulum Proteins:


Studies on the Structure and Distribution of

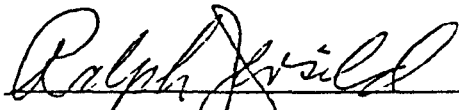
Calsequestrin and Phospholamban

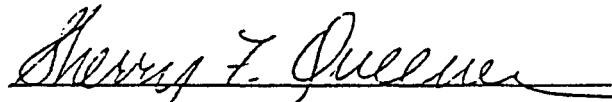
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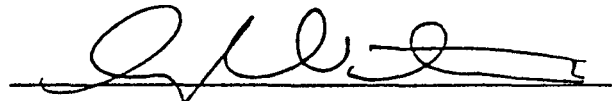
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Abstract

Complementary DNA clones of cardiac calsequestrin and phospholamban were isolated and novel isoforms of both of these sarcoplasmic reticulum proteins detected. The complete amino acid sequence of cardiac calsequestrin, a calcium binding protein of junctional sarcoplasmic reticulum, was deduced from the complementary DNA sequence. Cardiac calsequestrin was found to be an acidic protein of 410 amino acids, with a canonical 19 amino acid signal peptide sequence and an unusual carboxyl terminus, where 28 of the final 37 residues are acidic. The mature protein has 391 amino acids, two putative glycosylation sites and an unglycosylated molecular weight of 45,296. Primary and secondary structure analyses reveal the carboxyl half of cardiac calsequestrin is probably responsible for most of the calcium binding. Calsequestrin may bind calcium by acting as a charged domain rather than by presenting repetitive single binding sites. RNA coding for a second cardiac calsequestrin isoform which has not been previously described was detected and appeared to arise from alternative splicing. Cardiac calsequestrin isoforms were found to be expressed in slow skeletal muscle but not in fast skeletal muscle. Multiple cardiac calsequestrin-like sequences occur in the genome; however, there is only a single copy of the gene for the isolated calsequestrin clones.

Clones for phospholamban, an intrinsic protein of the sarcoplasmic reticulum, were isolated from dog heart, rat heart and rat soleus cDNA libraries. Two novel phospholamban isoforms were found in dog heart. In rat, three isoforms were detected. These clones are the first demonstration of distinct phospholamban isoforms.

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