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Localization of Adenylate Cyclase in Unfixed Sections of Cardiac Muscle

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Previous investigators have shown that prefixation and lead staining completely inhibit the activity of adenylate cyclase. Lead has also been shown to stimulate the nonenzymatic hydrolysis of AMP-PMP (the substrate for adenylate cyclase) after 30 min incubation. The present studies were performed to determine if the omission of prefixation would provide a better method for localizing adenylate cyclase in cardiac muscle. These studies were also performed to determine the effect of short incubation on the lead-induced nonenzymatic hydrolysis of AMP-PNP. In prefixed sections the reaction product was diffusely localized over the section. However, in unfixed sections the reaction product appeared only on the sarcolemma and sarcotubule system. Results are presented showing that short incubation (i.e., 5 min) prevents the nonenzymatic hydrolysis of AMP-PMP by lead. In biochemical studies lead (10^{-3} M) was shown to completely inhibit the activity of this enzyme. However, in the presence of 4 micrograms phosphatidylinositol, lead inhibition of this enzyme was reduced to 50% of the control value. Based on this observation, it is suggested that approximately 50% of adenylate cyclase is present in sections of cardiac muscle exposed to 2×10^{-3} M lead, which is presumably enough activity for demonstration of adenylate cyclase activity.

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