A BIOSENSOR TECHNOLOGY TO INVESTIGATE DRUG INTERACTIONS WITH MEMBRANE TRANSPORTERS

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The sarcoplasmic reticulum (SR) Ca-ATPase is a membrane transporter found in the SR of muscle cells [1]. This enzyme, which is a prominent member of the P-type ATPase family, hydrolyzes ATP in order to transport two Ca^{2+} ions against their electrochemical potential gradient from the cytoplasm to the lumen of the SR.
Charge transport in SR Ca-ATPase and related P-type ATPases has been extensively investigated in our laboratory using a biosensor technology, based on a solid supported membrane (SSM) [2]. The SSM consists of a hybrid alkanethiol/phospholipid bilayer supported by a gold electrode, and it represents a convenient model system for a biological membrane. Proteoliposomes or native membranes (vesicles or fragments) incorporating the transport protein can be adsorbed on a SSM and activated by a rapid substrate concentration jump. The substrate jump induces charge movement within the transport protein, resulting in a current transient which can be detected in the external circuit [2,3]. Therefore, the SSM serves two purposes at once, i.e. offering an adhesive surface to the adsorbed membrane entities and functioning as a transducer of a biosensor system.
Recently, our attention has been focused on the interactions of SR Ca-ATPase with molecules of pharmacological interest [4,5]. By combining biochemical and electrical measurements, we compared the effects of various compounds demonstrating different degrees of potency and specificity. We demonstrated interference of selected molecules with the Ca-ATPase transport cycle and we determined half-maximal inhibitory concentrations (IC_{50}) ranging from nM to μM values.
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