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Some properties of Ca^{2+} pumping ATPase in the symbiosome membrane from broad bean root nodules

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Ca^{2+} -pumping ATPase earlier identified by us in the symbiosome membrane (SM) of broad bean root nodules was functionally characterized by following its catalytic and transport activities. In the experiments with Ca probes arsenazo III and chlorotetracycline, the initial Ca^{2+} pumping rate was shown to achieve optimum at pH 7.2 and to significantly and only slightly decline in the vesicles SM and symbiosomes, respectively, at extreme chosen pH values, 6.0 and 8.0. It was established that the Ca^{2+} -ATPase is capable of utilizing not only ATP for fueling the Ca^{2+} transport through the SM but also other nucleotide triphosphates but with less efficiency, with its affinity for MgATP ($K_{0.5}$ for MgATP) is ~ 0.1 mM. In the reaction medium without added calcium, the rate of the Ca^{2+} -pumping was found to be markedly stimulated by exogenous calmodulin, a well-known intracellular Ca^{2+} -sensor. Based on the characteristics of Ca^{2+} -dependent ATP/ITP-hydrolysis by symbiosomes using the Ca-EGTA buffer system, an affinity of the Ca^{2+} -ATPase for Ca^{2+} ($K_{0.5}$ for free Ca^{2+}) was shown to be ~ 0.1 μM . In addition, it was found that the rate of Ca^{2+} -dependent ITP hydrolysis by symbiosomes in the presence of KCl in the reaction medium was significantly inhibited by nigericin indicating sensitivity of this process to symbiosome interior alkalinization. This finding and also revealed earlier insensitivity of the Ca^{2+} -ATPase to SM electric polarization are consistent with the proposal that this enzyme uses H^+ as a counterion to facilitate transmembrane calcium translocation. Taken together, these results allow us to refer the SM Ca^{2+} -pump to IIB type Ca^{2+} -ATPases family.

Biography

Krylova V.V. is senior science worker of Timiryazev Institute of Plant Physiology, Russian Academy of Sciences, Moscow. She made her Ph.D. at the problem Ca^{2+} transport across peribacteroid membrane of broad bean and lupine root nodules. She is author of more than 40 publications, 16 from these in reputed journals.