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金属离子对GTP结合蛋白 Cdc42Hs的内源性 GTP酶活性的影响

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不同的金属离子会对GTP结合蛋白Cdc42Hs的内源性GTP水解酶活性产生不同的影响。相对于生理条件下的辅基 Mg^{2+} 而言, Mn^{2+} 对Cdc42Hs酶活力有所激活,表现在饱和浓度时,实验曲线指数项的表观速率常数kobs有2倍左右的提高,而其稳态反应速度要低于 Mg^{2+} 。 Mg^{2+} 和 Mn^{2+} 的实验曲线在本质上没有什么差别,都是一个指数项和一个一次项的叠加,说明 Mn^{2+} 和 Mg^{2+} 以相似的机制结合于Cdc42Hs。在 Ca^{2+} 存在时,实验曲线无明显的指数项出现, Ca^{2+} 的存在仅使稳态反应速度有所降低,说明 Ca^{2+} 以不同于 Mg^{2+} 和 Mn^{2+} 的机理与Cdc42Hs结合。随着 Mg^{2+} 和 Mn^{2+} 离子浓度的增大,指数项的表观速率常数 k_{obs} 逐步升高,稳态反应速度 v_s 逐渐降低。进一步的动力学模型分析得到了这一反应的微观动力学常数和 Mg^{2+} 、 Mn^{2+} 与蛋白的结合常数。

EFFECTS OF METAL IONS ON THE INTRINSIC GTP-HYDROLYSIS ACTIVITIES OF SMALL GTPase Cdc42Hs

Different metal ions have different effects on the intrinsic GTP-hydrolysis activity of Cdc42Hs. Compared with Mg^{2+} , the physiological cofactor, Mn^{2+} has a slight activation effect on GTP-hydrolysis activity. The kobs of the exponential phase of the experiment curve of Mn^{2+} is two-times higher than that of Mg^{2+} under saturated condition. And the velocity of the steady state is lower than that of Mg^{2+} . Essentially, there is no difference between the experiment curves of Mn^{2+} and Mg^{2+} . Both of them have an exponential phase and linear phase, which indicate that Mn^{2+} and Mg^{2+} adopt the same mechanism in binding to Cdc42Hs. In the case of Ca^{2+} , there is no detectable exponential phase in the experiment curve. The presence of Ca^{2+} only slows down the velocity of the steady state. This indicates that the binding mechanism of Ca^{2+} to Cdc42Hs is different from that of Mn^{2+} and Mg^{2+} . With the increasing of Mn^{2+} and Mg^{2+} concentration, the kobs of the exponential phase increase, and the velocity of the steady state decrease. A detail kinetic analysis deduces the microscopic kinetic constants of the hydrolysis reaction and the disassociate constants of the metal ions with the protein.

关键词

金属离子(Metal ions); GTP结合蛋白(GTP-binding protein); Cdc42Hs; GTP水解酶活性(GTP-hydrolysis activity); 结合常数(Disassociate constants)