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Misoprostol and the Sildenafil analog (PHAR-0099048) Modulate Cellular Efflux of cAMP and cGMP Differently

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ABSTRACT

In the present study we have characterized ATP-dependent transport of cAMP and cGMP in physiological, but also supraphysiological concentrations. The uptake into inside-out vesicles from human erythrocytes could be dissected into two components with high and low affinity. The respective K_m-values were 30.8 ± 5.2 and 352 ± 26 μM for cAMP and 2.6 ± 0.4 and 260 ± 15 μM for cGMP. The two cyclic nucleotides were unable to mutually inhibit cellular efflux for concentrations up to about 100 μM. At higher concentrations the inhibition curve showed a steep fall. The IC_{50}-value for cAMP reduction of high affinity [^{3}H]-cGMP transport was 695 ± 9 μM. The respective value for cGMP inhibition of [^{3}H]-cAMP efflux was 284 ± 20 μM. These observations are compatible with two selective high affinity transport systems. Other endogenous substances such as prostaglandins did not discriminate between cyclic nucleotide transport. The IC_{50} values for inhibition of [^{3}H]-cAMP and [^{3}H]-cGMP were 4.1 and 4.2 μM for PGE_1, 2.7 and 4.4 μM for PGE_2, respectively. However, the prostaglandin analog misoprostol discriminated distinctly between cAMP and cGMP transport with respective IC_{50}-values of 4.5 and 24 μM. The assumption that the specific PDE5-inhibitor sildenafil could distinguish between the two cyclic nucleotides was disproved with respective IC_{50} values of 3.8 and 2.9 μM for inhibition of [^{3}H]-cAMP and [^{3}H]-cGMP, respectively. However, at least one sildenafil analog (PHAR0099048) showed a clear difference with respective IC_{50} values of 2.0 and 0.52 μM. The other tested sildenafil analogs showed no or minor ability to discriminate with IC_{50} values of 0.16 and 0.17 μM for IS-39213, and 0.35 and 0.16 μM for IS-60049, respectively. In agreement with previous reports, the present study shows that proteins responsible for cyclic nucleotide transport are multiorganic anion pumps. However, the observation that drug analogs may discriminate between these two efflux systems makes them potential drug targets.

KEYWORDS

ABC-Transporters; cAMP; cGMP; Misoprostol; Sildenafil

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