923/E2

Molecular investigation of the synergetic activity between Na^+ /K^+ carrier ionophore antibiotics and fluoride

S.A.D.N. Dias, S. Divyasorubini and G. N. Silva*

Department of Chemistry, University of Colombo, Colombo 03, Sri Lanka

The antibiotic resistance and host toxicity has limited the clinical use of many antibiotics. For example, membrane destabilizing Na^+/K^+ carrier ionophores such as valinomycin and monensin, exert their antibiotic activity by conducting ions through the cell membrane but can cause host toxicity by perturbing intracellular ion homeostasis. Nonetheless, valinomycin has exhibited remarkable inhibitory effects against coronaviruses, including SARS-CoV-2, the causative agent of the current global pandemic COVID-19. Therefore, translating such compounds to clinical settings requires them to remain non-toxic to the host. Fluoride is a toxic anion that has both antimicrobial and virucidal properties. However, the cellular uptake of fluoride is limited due to the bacterial cell envelope. Accordingly, compounds that enhance the permeability of cell membranes should induce strong fluoride toxicity in pathogens by enhancing fluoride uptake. Therefore, combination regimens such as ionophore antibiotics and fluoride may increase treatment efficacy, while lowering the doses of host-toxic antibiotics. In this study, we demonstrate that fluoride significantly enhances the antibacterial activity of the carrier ionophore antibiotics; valinomycin and monensin. Cell growth assays were separately performed by incubating the Gram-positive bacteria (*Bacillus subtilis, Bacillus megaterium and S. aureus*) at 37 °C with monensin (0.2 µg/ml) and valinomycin (50 µg/ml) with varying concentrations of NaF. Then, OD_{600} of the cultures were measured after 16 hours of incubation. Our results demonstrate that the potency of monensin and valinomycin was increased by 5-fold and 2-fold, respectively, in the presence of sub-inhibitory concentrations of NaF (~100 mM). In contrast, neither of the antibiotics exert any synergism with fluoride against the Gram-negative bacterium *E. coli.* Overall, it can be suggested that the two ionophore antibiotics may increase the cellular uptake of fluoride to exert synergistic bacterial growth inhibition by enhancing intracellular fluoride toxicity. This study provides new insights to design novel and effective therapeutic combination regimens for deleterious diseases that have plagued humans.

Keywords: Valinomycin, monensin, fluoride, Na^+/K^+ carrier ionophore, Gram-positive bacteria

E-mail: gayathris@chem.cmb.ac.lk