Binding of aminoglycosidic antibiotics to the oligonucleotide A-site model. Brownian dynamics simulations.

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AIMS: The encounter of four different aminoglycosidic antibiotics with the ribosomal A-site is simulated with Brownian dynamics methodology. We analyze the influence of structural, electrostatic and hydrodynamic properties of antibiotics on the kinetics and mechanism of their encounter with RNA. Diffusion limited rates of association are computed and their dependence on ionic strength is examined.

AMINOGLYCOSIDES: Aminoglycosidic antibiotics are a family of antibacterial drugs. Most aminoglycosides block proper peptide synthesis by binding to the prokaryotic tRNA decoding A-site of the 16S RNA in the 30S subunit. Aminoglycosides suffer from moderate affinity, inadequate specificity and can be toxic. Moreover, bacterial resistance limits their effectiveness in medical therapy. Therefore, we need to understand their binding mechanism in order to be able to improve their selectivity and efficiency. Studied aminoglycosides belong to the same family of sugar derivatives but differ in the number of rings, amine and hydroxyl groups, as well as the total charge. All antibiotics are positively charged at physiological pH and their diffusion toward RNA is electrostatically driven.

BROWNIAN DYNAMICS: A three dimensional ligand, composed of spherically symmetric subunits diffuses to a fixed receptor.

\[
\begin{align*}
    r_{i}^{n+1} &= r_{i}^{n} + \sum_{j} \frac{\Delta t}{k_{B} T} D_{ij}^{n} F_{j}^{n} + R_{i}(\Delta t) \\
\end{align*}
\]

Ligand is subjected to external forces, derived from the Poisson-Boltzmann equation. Probability of reaction and association rate constant can be computed from ensemble of Brownian dynamics trajectories.

ASSOCIATION RATES: The values of association rate constants obtained in this work are of the order of $10^{-10}[\text{Ms}]^{-1}$. Largest values are observed for neamine. Smallest rates are those obtained for paromomycin and neomycin, both composed of four rings. Difference between their kinetics, resulting from higher total charge of neomycin, is visible only at low ionic strengths.

ASSOCIATION MECHANISM: Based on Brownian dynamics trajectories we calculated densities of paromomycin diffusing in the potential generated by:
- the naked RNA fragment (green) and
- the RNA fragment with one of the A-sites permanently occupied by another paromomycin (yellow).

CONCLUSIONS: The behavior of ligands observed in BD trajectories suggests that the binding of antibiotics to the A-site can be accomplished in two ways: the ligand either finds the binding site directly or, due to the highly negative potential of the RNA fragment, associates on the oligonucleotide surface and then slides along the groove between backbone phosphate groups, in a manner of one-dimensional diffusion, until it finds the binding pocket. The kinetics of the RNA oligonucleotide/antibiotic complex formation has not been studied previously neither with experimental nor computational methods and we are not able to compare our results with those obtained with another method.

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