Antibacterial resistance via GyrB24 targeting

Abstract

In nature, four mutations confer resistance to coumarins in Escherichia coli strains: Arg 136 to Cys, His or Ser and Gly-164 to Val. All of these mutations are located at the ATP-binding site for the B subunit of the gyrase enzyme. GyrB proteins with these mutations lose their enzymatic activity. Molecular visualizations showed that Arg-136 in gyrB24 is hydrogen bounded through its guanidinium group to the two oxygen atoms in the Coumarin ring. Planning of new compounds with more profound antibiotic activities includes extending the alkyl group at the 5 position of the noviose sugar for the Coumarin. On the other hand, more studies are needed to assess the molecular interactions between coumarins and the host cells to identify specific moieties involved in allergy (jaundice) reactions evoked by the antibiotics.

Abbreviations: UPEC, uropathogenic escherichia coli

Introduction

In Escherichia coli, the gyrase enzyme is composed of two subunits: A and B. The A subunit (97k Da) interacts and relieves DNA super coils through its active site (tyrosine) during DNA replication. The B subunit contains the ATP as active site required to release the energy in the attached ATP molecule and provides the free energy to the reaction (DNA super coiling) accomplished by the gyrase enzyme. Amino coumarins/coumarins such as novobiocin and coumermycin A1 are associated with a 24-kDa (residues 2-220) sub-domain in the B subunit of the gyrase enzyme known as amino terminal. This association overlaps the ATP as site in gyro Band prevents further binding of ATP molecules. However, the poor activity of aminocoumarinins inhibitors for Gram-negative bacteria, their cytotoxic effects on mammalian cells and low water solubility reduced their use in clinical practice. On the other hand, new studies are investigating the possibility of emulating the mode of action of coumarins using new molecules with less toxic effects and better inhibitory reactions. In nature, four mutations confer resistance to coumarins in Escherichia coli strains: Arg-136 to Cys, His or Ser and Gly-164 to Val. All of these mutations are located at the ATP-binding site for the B subunit. GyrB proteins with these mutations lose their enzymatic activity. Other investigators introduced site-directed mutations at amino acids interacting with aminocoumarinsas. The mutations were able to reduce binding of protein with aminocoumarinsas but at the same time were unable to bind ATP as well. The introduced mutations were Asp73 to Asp and Asn46 to Asp and Leu in GyrB of E. coli spp. Other investigators introduced the following site-directed mutations in gyrB: Asp73 to Glu, Gly77 (to Ala and Ser), Ile78 (to Ala and Leu), and Thr165 (to Ala and Val). They observed that the E. coli mutants were resistant to novobiocin but with low clear loss of enzymatic activity. One important aspect noted during the study of these mutations regarding the size of the interacting molecule. Studies indicate that the number of produced mutations is remarkably linked to the size of the drug. For example, cyclothialidines (Antibiotic family with similar properties as aminocoumarinsas, targeting the ATP use site of gyrB, but usually are larger) are known to complex and overlap regions of the gyrase B subunit as aminocoumarinsas. However, the type of interaction made by cyclothialidines and coumermycin appears to be more profound with more contact areas on the gyrase B subunit than is the case with novobiocin. For example, investigators noted that a single mutation could confer resistance to high concentrations of novobiocin. While, a remarkable decrease in bacterial susceptibility to both cyclothialidines and coumermycin in is associated with double mutations at the ATPase site. The reference attributed the case above to the “bulkier molecular nature” of coumermycin and cyclothialidine compared to novobiocin. Effects of gyrase mutations on the expression of virulence genes in pathogenic strains

Due to the importance of gyrase for both DNA replication and gene expression, it is expected that the expression of certain core genes will be affected by the mutations that reduce the enzyme activity. Mutations in the gyrA gene of Uropathogenic Escherichia coli UPEC are known to cause a decrease in the virulence of pathogenic bacteria due to their effects on DNA super coiling and expression of several virulence factors. Such UPEC strains showed decreasing capacity to cause cystitis and pyelonephritis. However, no similar studies support a similar finding for mutations in gyrB and coumarin resistance.

Discussion

The catalytic ATPase site of gyrB is a 24 KDa fragment. This site is target for known coumarins. Four chemical groups are making the coumarin: L-noviose sugar, the 5-methylpyrrole-2-carboxylic acid, the 3-amino coumarin ring, and the 4-hydroxy-3-isoprenylbenzoyl moiety. Molecular structure analysis showed that Arg-136 in gyrB24 is hydrogen bounded through its guanidinium group to the two oxygen atoms in the Coumarin ring. Mutations in the gyrA gene of Uropathogenic Escherichia coli UPEC are known to cause a decrease in the virulence of pathogenic bacteria due to their effects on DNA super coiling and expression of several virulence factors. Such UPEC strains showed decreasing capacity to cause cystitis and pyelonephritis. However, no similar studies support a similar finding for mutations in gyrB and coumarin resistance.
the noviose sugar are contributing to the hydrophobic interactions with the following residues in the ATPase site: Ile 78 and Ile90. The remaining 5-methylpyrrole ring is also involved in hydrophobic pocket formations within gyrB24 with the following interaction points Val43, Ala47, Val71, Ile78, Val120, and Val167. The above information represent the importance of key residues in the hydrophobic interactions between coumarins (inhibitors) and the 24kDa ATPase site and that in the absence of the antibiotic these hydrophobic pockets are occupied with trapped water molecules. Understanding the molecular basis for the interactions between the antibiotics and their target sites would enable better adjustments to enhance activity and affinity of the antibiotics. From a literature point view it looks that more studies are needed to assess the molecular interactions between coumarins and the host cells to identify specific moieties involved in allergy (jaundice) reactions evoked by the antibiotics.

**Conclusion**

GyrB mutations rendering the bacterial cells resistant to aminocoumarins as also involved in deactivating the enzyme. The number of mutations introduced depends on the size of the molecule. Studies showed that water molecules surrounding the gyrB24 protein number of mutations introduced depends on the size of the molecule. Future design of studies showed that water molecules surrounding the gyrB24 protein and affinity of the antibiotics. From a literature point of view it looks that more studies are needed to assess the molecular interactions between coumarins and the 24kDa ATPase with the following residues in the ATPase site: Ile 78 and Ile90. The 24 kDa N-terminal sub-domain of the DNA gyrase B protein binds coumarin drugs. Molec. Microbiol. 1994;12(3):365–373.

**Acknowledgements**

None.

**Conflict of interest**

Author declares that there is no conflict of interest.

**References**
