Mechanisms of Multidrug-Resistance in *Pseudomonas aeruginosa*

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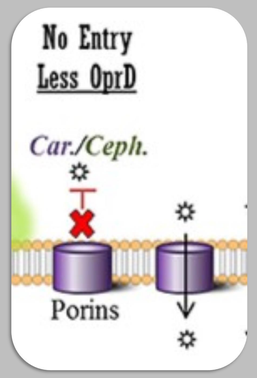
Characteristics & Epidemiology

Multidrug-resistant *Pseudomonas aeruginosa* is an opportunistic pathogen that is classified as a major threat to public health in the United States today (2) (3). This gram-negative bacillus bacteria is nosocomial and is transmitted through contact with contaminated particles in water and soil, as well as through medical devices and unclean hands (2). Patients in hospitals that are immunocompromised, use devices such as ventilators, have pre-existing illness, and have exposed wounds due to operations or burns are at the greatest risk of experiencing serious illness (2). The accumulation of several mechanisms to block antibiotic interactions with the bacteria is a growing problem in healthcare settings.

Outer-membrane & Porins

Gram-negative bacteria several protective layers, including the cellular membrane, cell wall, and outer membrane (3). *P. aeruginosa* has specific porin proteins residing on its selectively permeable outer membrane that provide intrinsic antibiotic resistance. Mutations have also reduced the number of porins (OprD), resulting in strains that are resistant to antibiotics such as carbapenems and cephalosporins, shown in Figure 3 (5).

Figure 3: Under expression of porins on the outer membrane (5)
Transmembrane proteins called porins act as a pathway for antibiotics into the cell. Mutations have caused an under expression of these proteins in *P. aeruginosa* , leading to antibiotic resistance against carbapenems (Car.) and cephalosporins (Ceph.).



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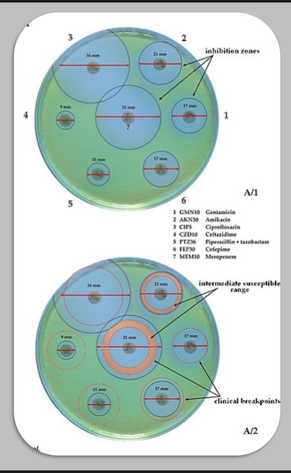
Resistance Overview

Through years of mutations and adaptation, the bacterium has acquired multiple mechanisms that prevent antibiotics from inhibiting its proliferation, shown in Figure 1 (5)..

These mechanisms include:

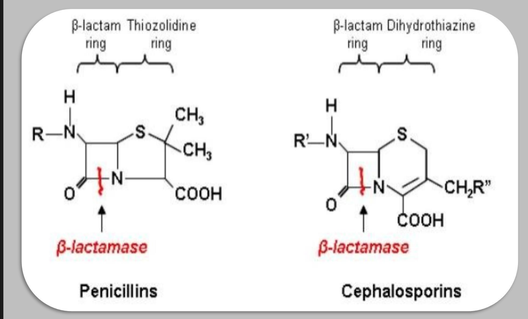
- Outer-membrane selective permeability
- Antibiotic-inhibiting enzymes
- Biofilm creation
- Efflux pumps
- Horizontal gene transfer

Figure 1: *P. aeruginosa* antibiotic resistance (11)
Discs containing various antibiotics were added to a petri dish containing *P. aeruginosa*. The antibiotics were successful in preventing growth of the bacteria or failed to do so to varying degrees. The circles, called inhibition zones, with shorter diameters (4 and 5) show high levels of antibiotic resistance; conversely, those with large diameters (3 and 7), show little resistance to the antibiotics.



Antibiotic-Inhibiting Enzymes

Figure 4: Breaking the β-lactam ring found in antibiotics (10)
The ring that forms a major portion of the antibiotic's molecular is broken by β-lactamases produced by the bacterial cell.

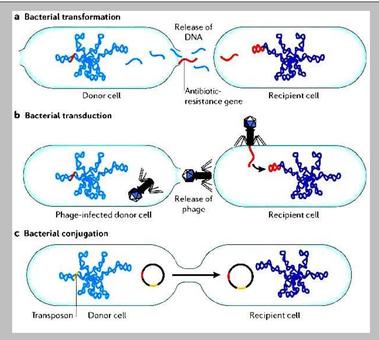


P. aeruginosa has genes that code for β-lactamases that can hinder antibiotics by breaking down the β-lactam ring present, rendering them inactive (5). Mutations have led to overproduction of this mechanism, leading to increased antibiotic inhibition (5). The antibiotics by penicillins, aminoglycosides, cephalosporins, and fluoroquinolones are restricted due to this mechanism because they are unable to prevent the bacterial cell from carrying out cell wall synthesis (3) .

Transferring of antibiotic resistance

Since the genetic information for several of the antibiotic resistance can be found in plasmids, it can easily be transferred to another bacterium, leading to more widespread antibiotic resistance (5). Specific adaptations such as overproduction of efflux pumps and creation of antibiotic-inhibiting enzymes can be passed on during gene transfer (3). The ability for resistance to be easily passed from one cell to another contributes to its dangerous nature.

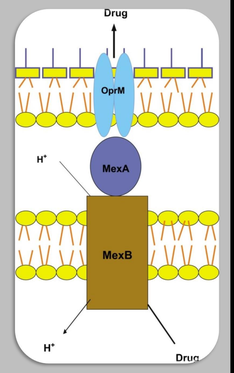
Figure 5: Horizontal gene transfer mechanisms (4)
Genes with resistance can be transferred between bacteria by transformation, transduction, and conjugation.



Efflux Pumps

Efflux pumps are trans-membrane proteins that expel antibiotics from bacterial cells (3). *P. aeruginosa* has multiple complexes containing multidrug efflux (Mex pumps) and an outer porins (Opr) that can identify and excrete a broad spectrum of antibiotics including aminoglycosides, penicillins, and fluoroquinolones classes (Figure 2) (1). Mutations in these proteins result in an overproduction of efflux transmembrane proteins, increasing the bacterium's antibiotic resistance (5).

Figure 2: Mechanism of Efflux Pumps (7)
The antibiotic that is found inside of the cell is expelled using a complex of proteins, including Mex pumps and outer porin (Opr) channels.



Biofilm Production

Biofilms are collections of cells anchored to a surface that prevent antibiotic interference and promote continued growth, especially in the human respiratory system (3). The many species that occupy these biofilms synchronize processes by quorum sensing, leading to a spread of resistance between the same species and a uniformity to the structure (3). The cells form a matrix that strengthen the colonies, posing a worrisome resistance for patients with respiratory illness such as cystic fibrosis (9).

Figure 6: Biofilm formation (7) (6)
Biofilms have four stages involving basic steps, including attachment, expansion, maturation, and dispersion. The many cells in addition to the matrix formed provide protection from antiibotics.