Effect of Lipopolysaccharide on Antibiotic Susceptibility in *A. Baumannii*

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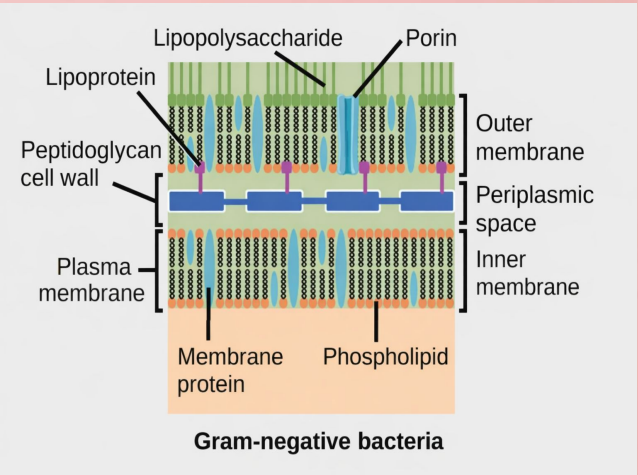
Research Question:

How does the absence of lipopolysaccharide affect antibiotic susceptibility in *A.baumannii?*

Background:

- *A. Baumannii* is a gram-negative critical priority pathogen.
- Gram-negative bacteria have a lipopolysaccharide (LPS) layer.
- LPS acts a permeability barrier.
- *A. Baumannii* can survive without LPS.
- This research is examining susceptibility differences between wild-type strains and mutant strains.
- Using three antibiotics: daptomycin, clindamycin, and colistin.



Gram-negative bacterial structure.
Source: OpenStax Biology

Background cont’d:

- Daptomycin, and clindamycin normally target gram-positive bacteria.
- May show increased activity on gram-negative bacteria when LPS is compromised
- Colistin acts on gram-negative bacteria.
- May show decreased activity when LPS is compromised.
- This research is necessary for exploring how bacterial cell structures influence antibiotic resistance.

Hypothesis:

- If wild-type (AB-84) and mutant (AB-84R) *A. Baumannii* strains are tested under identical conditions:
  - AB-84R will be more susceptible to daptomycin, and clindamycin since they lack LPS.
  - AB-84R will be more resistant to colistin because it binds directly to LPS.

Procedure:

Grew cultures of both strains overnight.

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Standardized suspensions (**0.5 McFarland**).

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Plated strains (Mueller-Hinton agar plates).

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Placed antibiotic E-test strips onto plates.

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Incubated **37 °C** for **18–24 hours.**

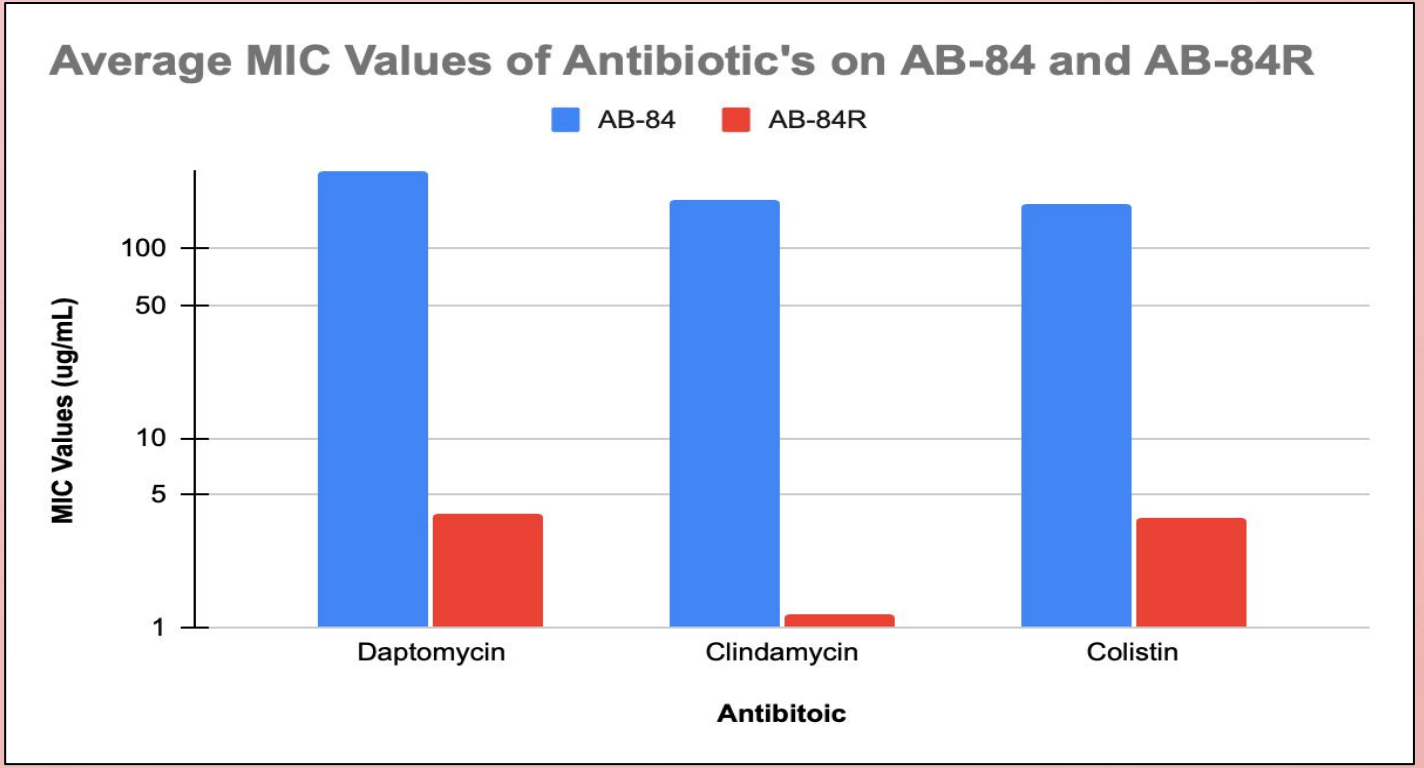
↓

Repeated across **6 trials.**

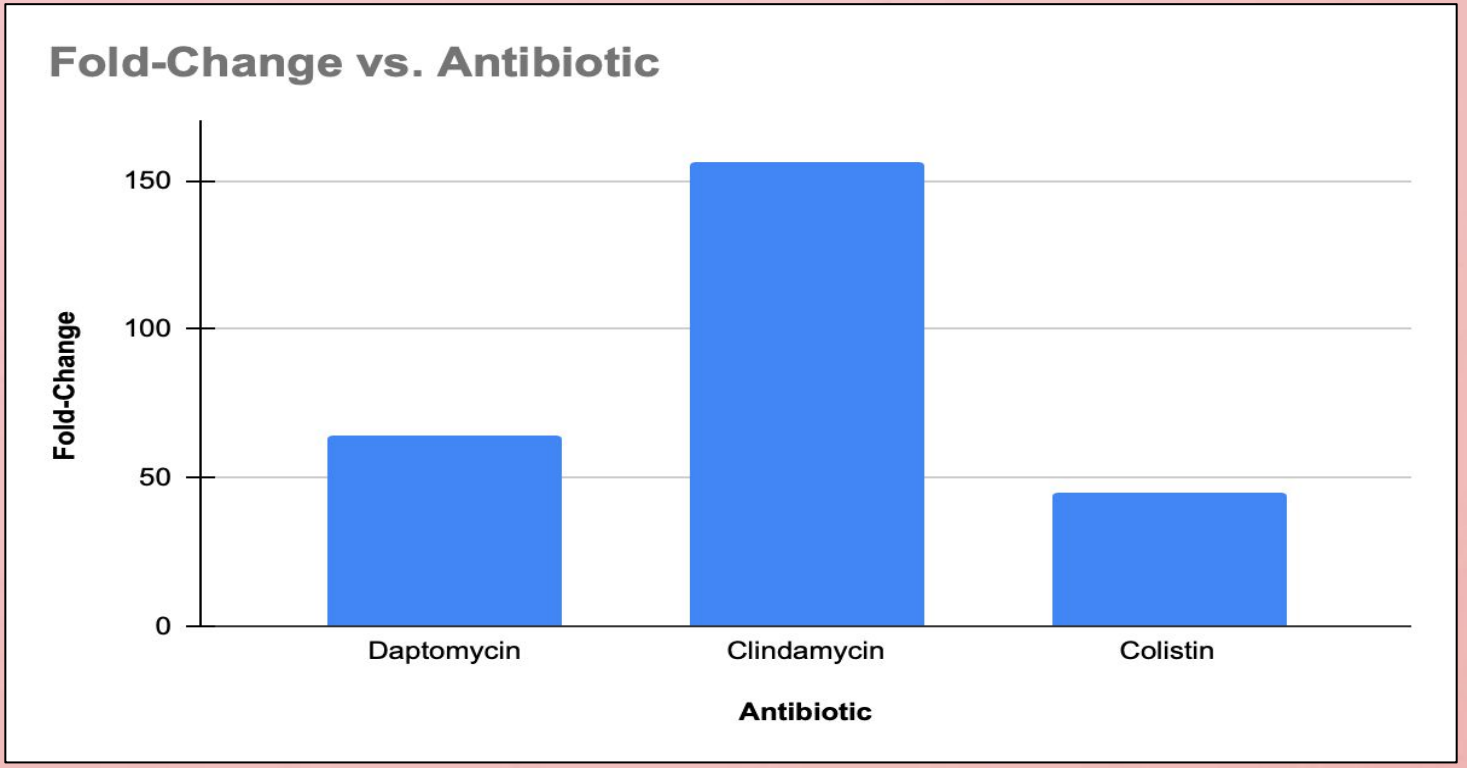
Results:

- MIC values were recorded for each antibiotic and averages were calculated in ug/mL.
- Daptomycin AB-84 mean was 256 ± 0.00, and it’s AB-84R was 4.00 ± 4.33.
- Clindamycin AB-84 mean was 181 ± 129, and it’s AB-84R mean was 1.16 ± 1.04.
- Colistin’s AB-84 mean was 171 ± 146, and it's AB-84R mean was 3.83 ± 3.62.

Graphs:


Average MIC Values of Antibiotic's on AB-84 and AB-84R
AB-84
AB-84R
MIC Values (ug/mL)
100
50
10
5
1
Daptomycin
Clindamycin
Colistin
Antibitoic

Graphs cont'd:



- Fold change represents susceptibility difference between wild-type and mutant strains.

Statistical Analysis:

Mean MIC values and Variability of AB-84 and AB-84R (ug/mL)

Antibiotic	AB-84 mean+SD	AB-84R mean+SD	Fold Change	P-Value
Daptomycin	256 ± 0.00	4.00 ± 4.33	64x	<0.001
Clindamycin	181 ± 129	1.16 ± 1.04	156x	0.002
Colistin	171 ± 146	3.83 ± 3.62	45x	0.026

Discussion:

- AB-84R was more susceptible to daptomycin and clindamycin than AB-84, supporting the hypothesis.
- Colistin results were inconsistent, and refuted the hypothesis in 4 of 6 trials.
- The absence of LPS resulted in a compromised outer membrane which influenced colistins functionality.
- AB-84 showed high variance, suggesting that LPS impacts susceptibility.
- AB-84R showed low variance, suggesting that the absence of LPS allows greater inhibition.
- Results emphasize how bacterial structures contribute to intrinsic resistance.

Conclusion:

- AB-84R strains were more susceptible to daptomycin, and clindamycin across all trials.
- Supports the hypothesis that the absence of LPS leads to increased inhibition of gram-positive antibiotics.
- Trials one and four of colston supported the hypothesis that AB-84R would be more resistant, while all other trials refuted this.
- These other trials suggest that membrane stability has greater impact on functionality than the absence of LPS.
- This research supports developing treatments against critical-priority pathogens like *A. Baumannii.*
- Future pursuits should investigate how manipulating LPS could potentially improve antimicrobal treatments.