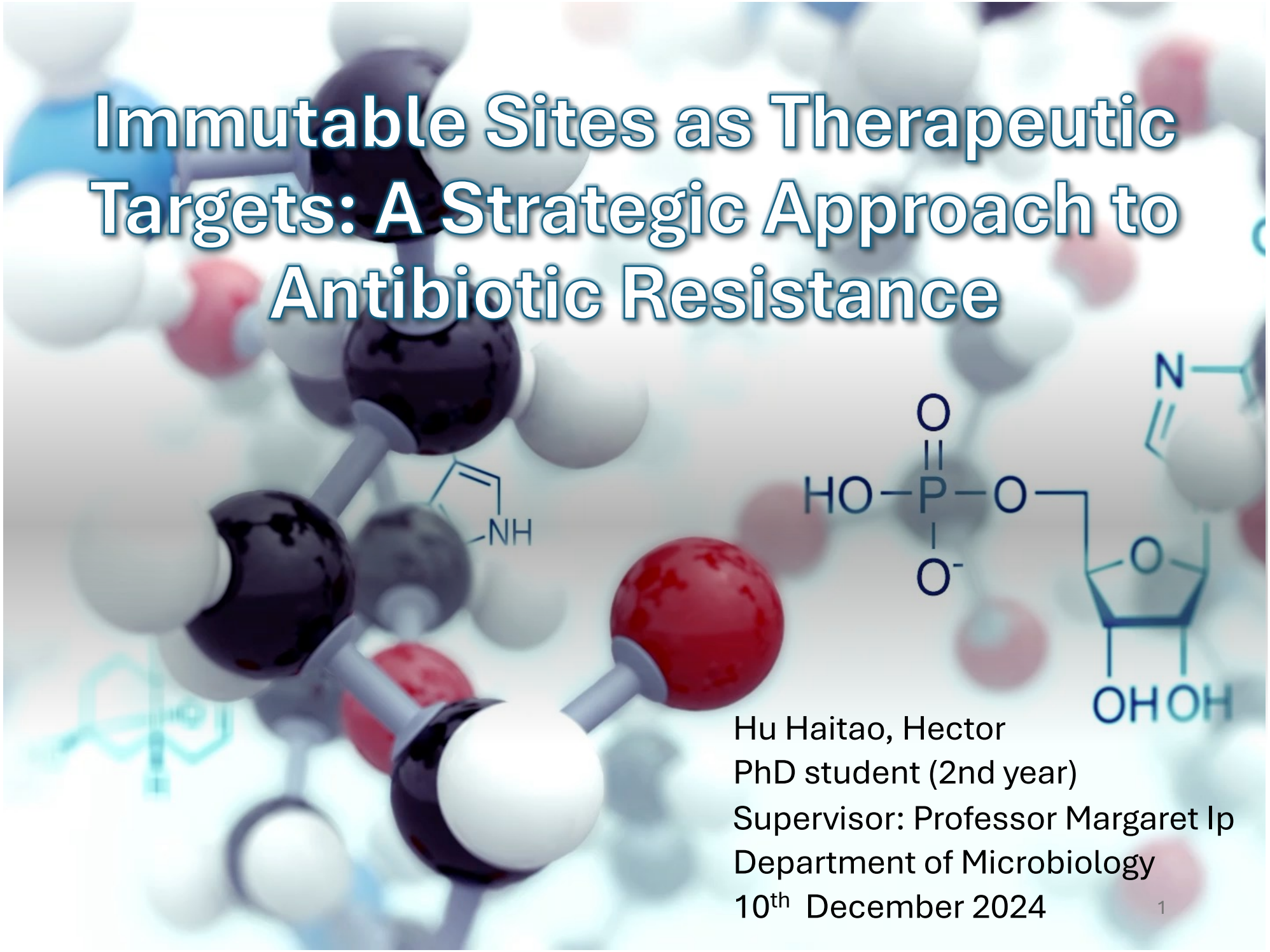
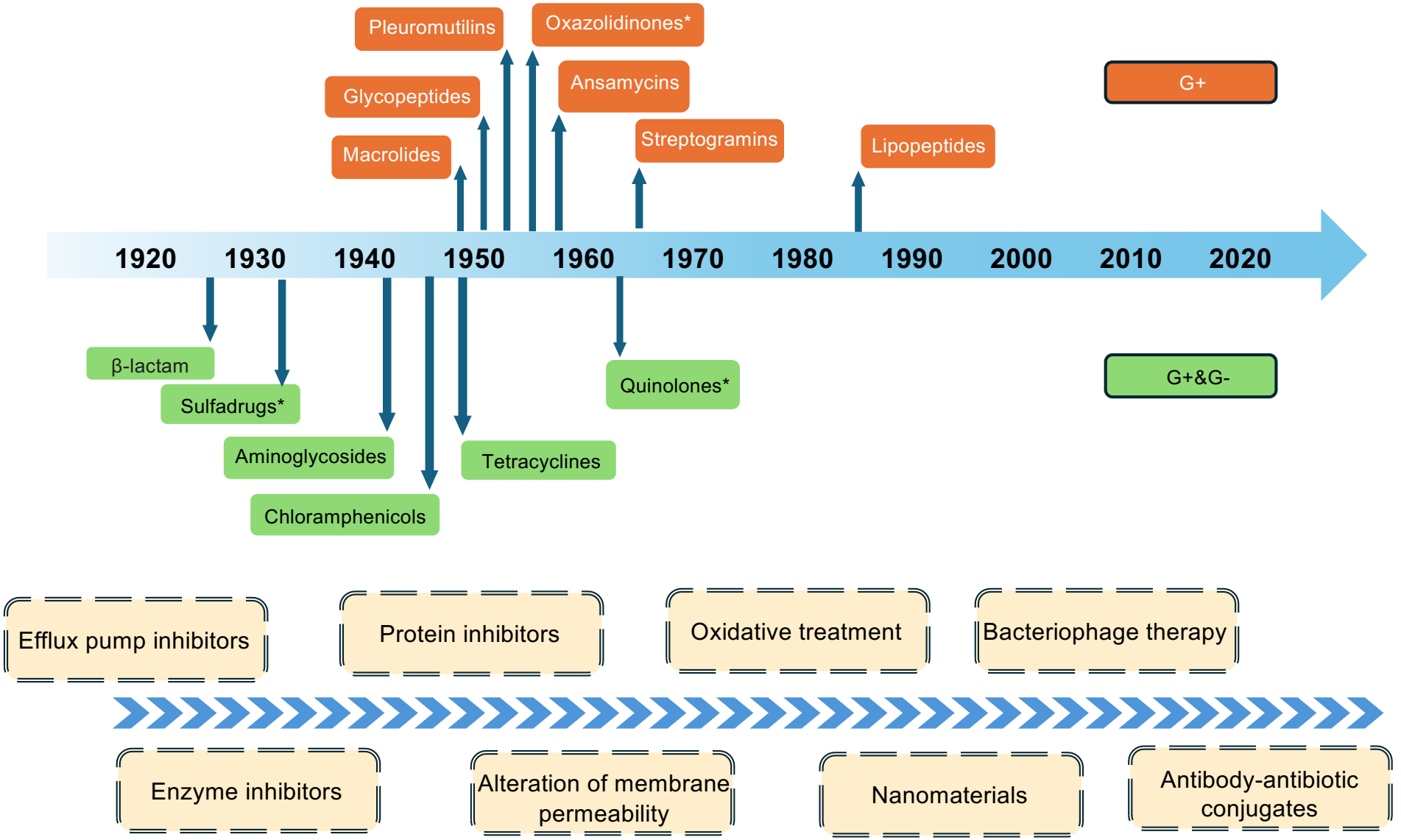
Immutable Sites as Therapeutic Targets: A Strategic Approach to Antibiotic Resistance



Hu Haitao, Hector
PhD student (2nd year)
Supervisor: Professor Margaret Ip
Department of Microbiology
10th December 2024

ANTIBIOTIC DISCOVERY



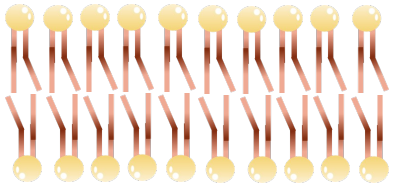
The timeline of clinically used antibiotic discovery and overview of mechanism-guided strategies for combating antibiotic resistance. (*) Denotes synthetic compounds.

(Biochemistry, 2020; World J Microbiol Biotechnol 2024)

ANTIBIOTIC CLASSIFICATION

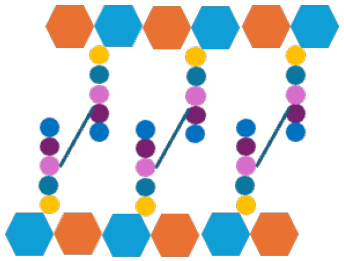
Cell envelope targeting

Membrane function



Gramicidin, cyclic
lipopeptides(G+), polymyxins

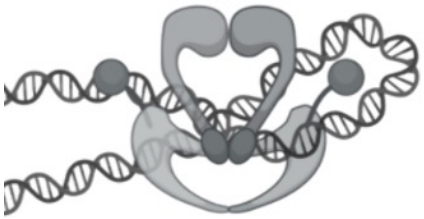
Peptidoglycan synthesis



β-lactams/β-lactamase inhibitors,
glycopeptides(G+)

Cytosolic steps targeting

DNA replication



Fluoroquinolone,
novobiocin

RNA synthesis

Ansamycin,
tiacumicin(G+)

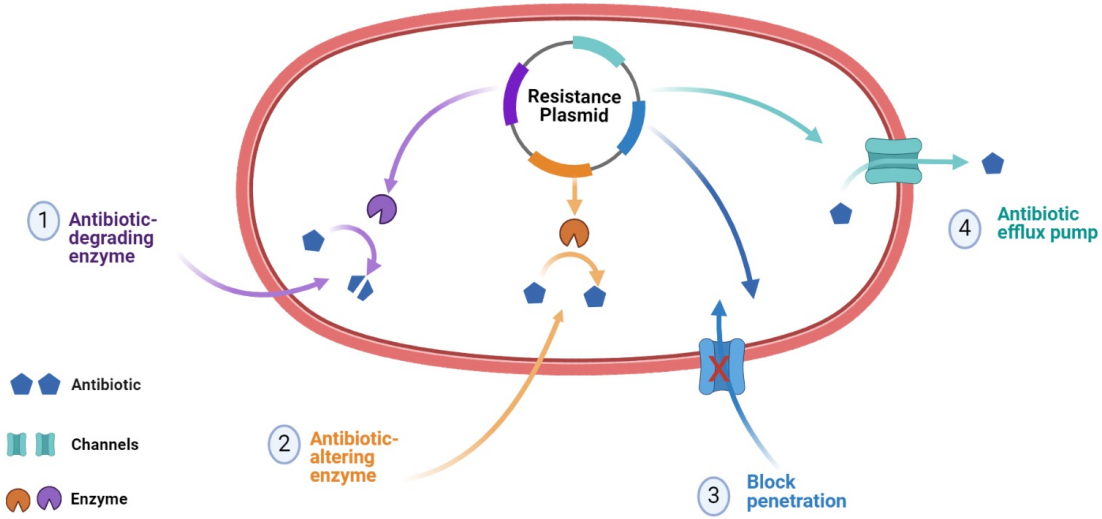
Protein synthesis

Aminoglycosides, oxazolidinones
tetracyclines, phenicols, streptogramin(G+)

(Microbiol Spectr, 2016; Ann Pharmacother, 2015)

Antimicrobial Resistance(AMR)

- An increasing resistance to frontline lifesaving antibiotics that undermines the ability to treat common and serious infectious disease
- Cause **700,000** deaths every year
- Increase to 10 million deaths a year by 2050



WHO reports widespread overuse of antibiotics in patients hospitalized with COVID-19

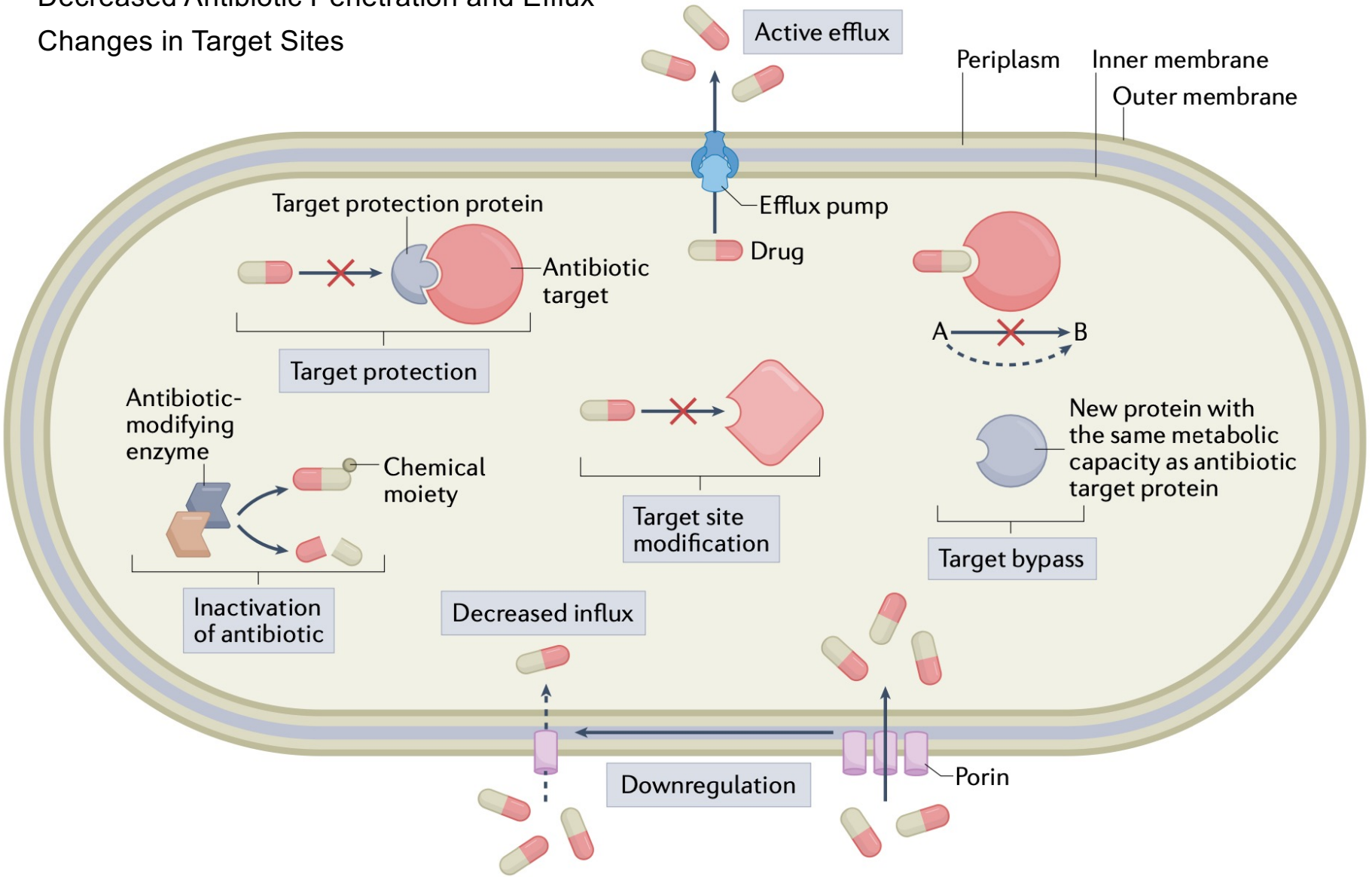
26 April 2024 | News release | Geneva |Reading time: 2 min (501 words)

New evidence from the World Health Organization (WHO) shows the extensive overuse of antibiotics during COVID-19 pandemic worldwide, which may have exacerbated "silent" spread of antimicrobial resistance (AMR).

(Usman Abubakar, 2022; Jeon, K. ,2022; Suriya Rehman,2023)

MECHANISTIC BASIS OF ANTIBIOTIC RESISTANCE

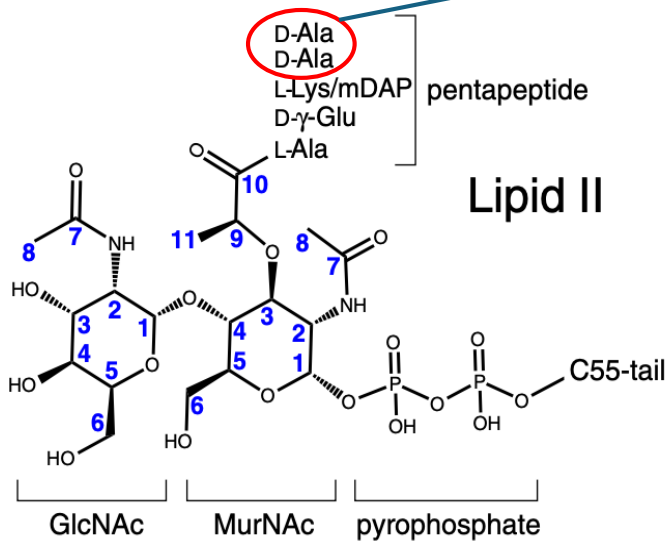
- Modifications of the Antibiotic Molecule
- Decreased Antibiotic Penetration and Efflux
- Changes in Target Sites



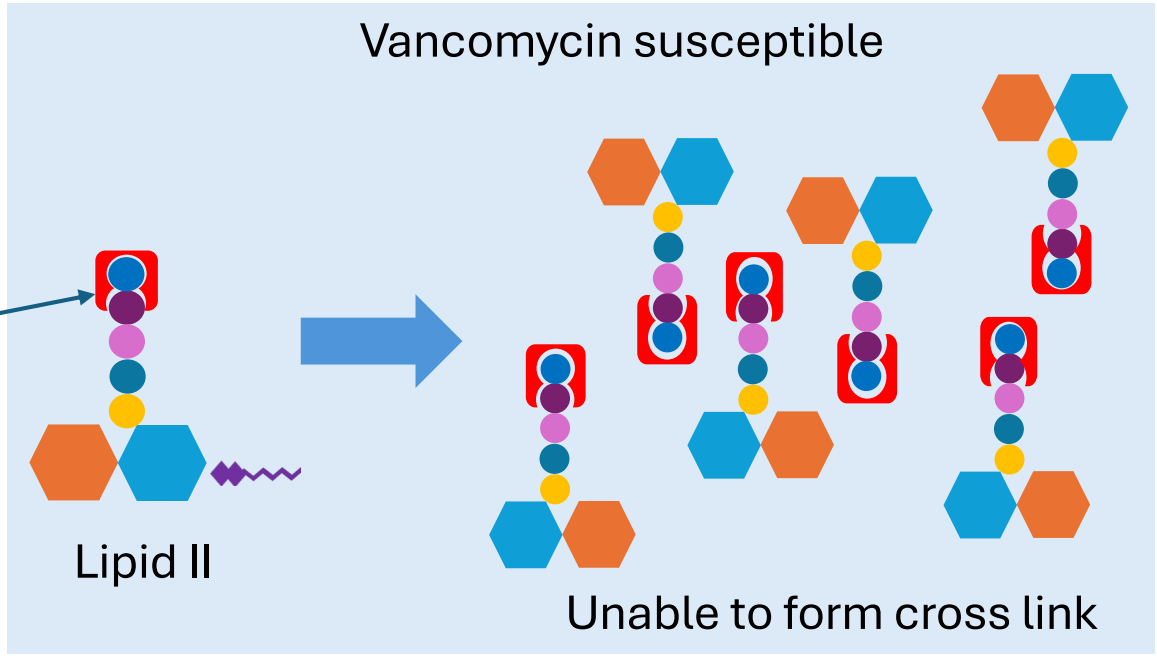
(Nat Rev Microbiol, 2023)

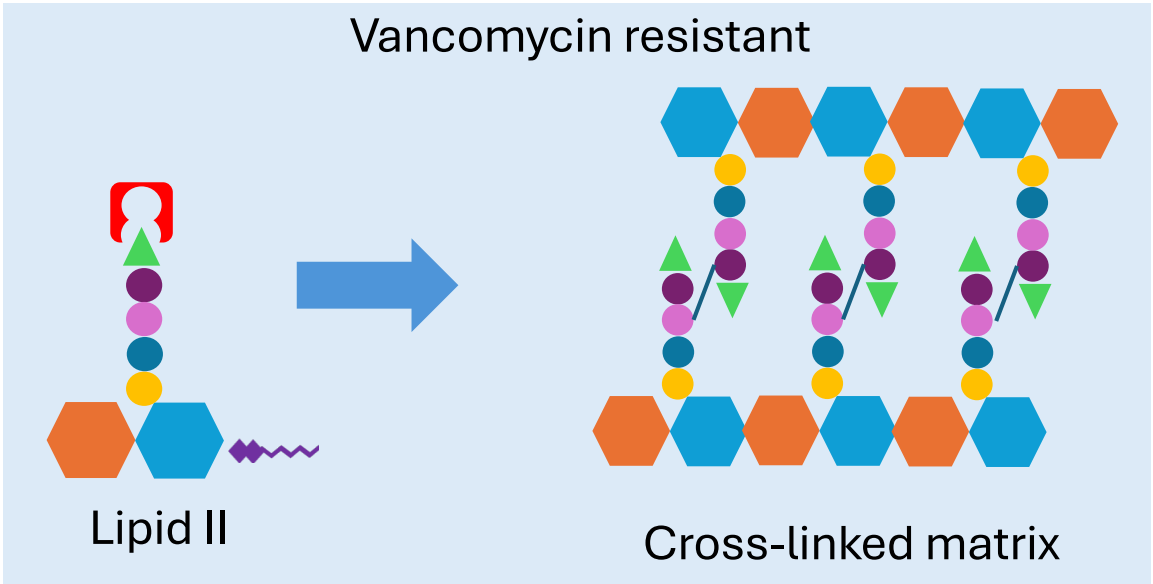
CONTENTS

Vancomycin binding scheme



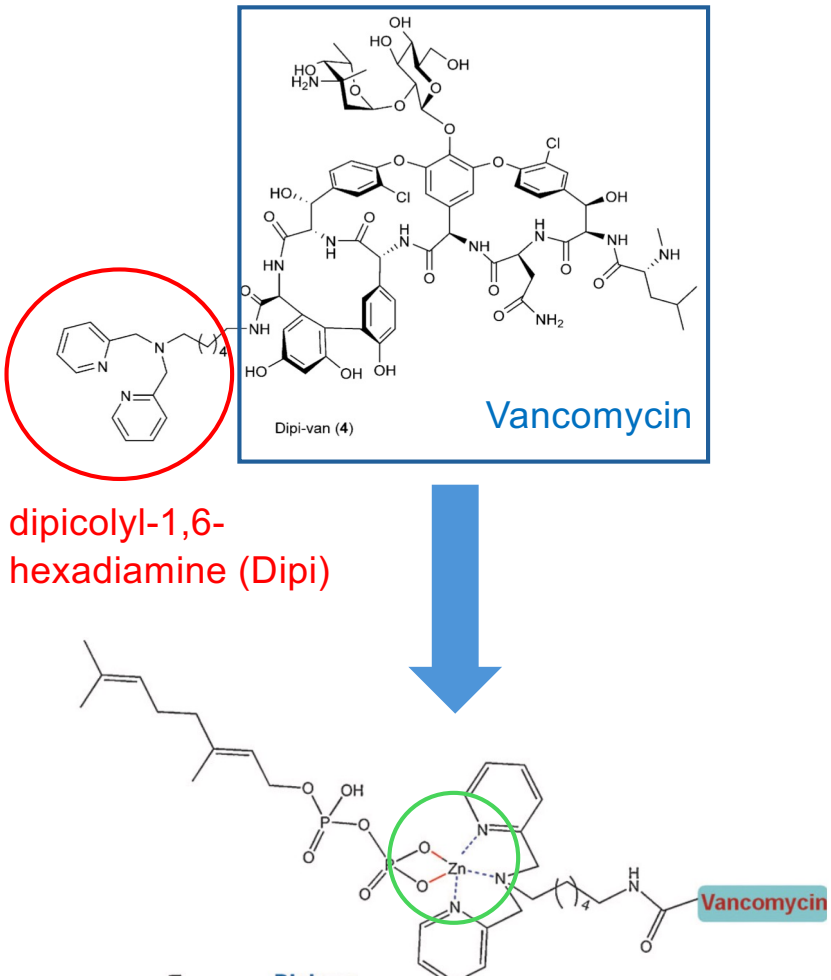
D-alanine→D-lactate

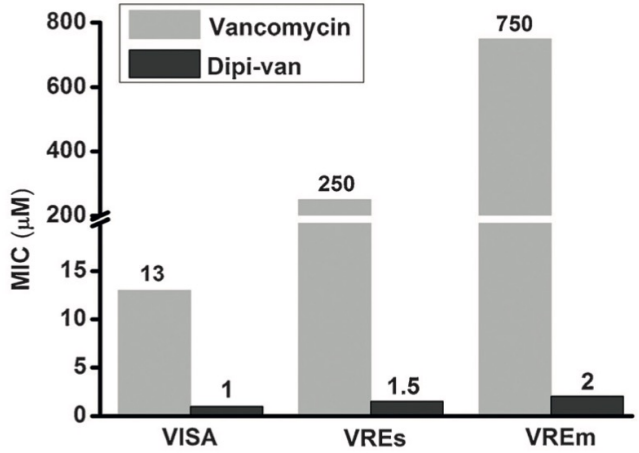


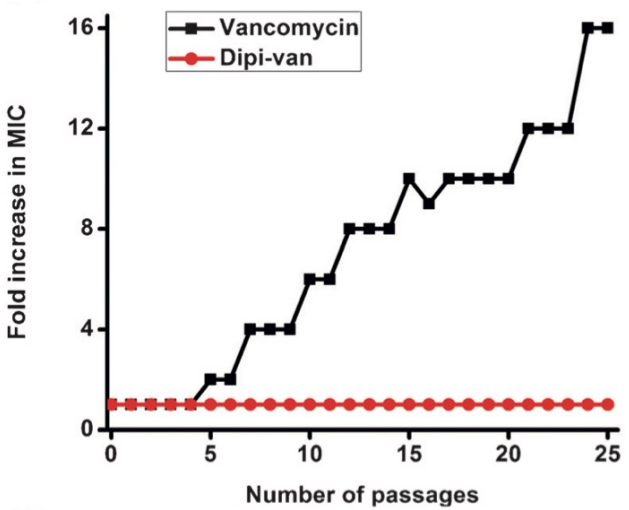


Resistance evasive antibiotics

Lipid II-caging vancomycin conjugates





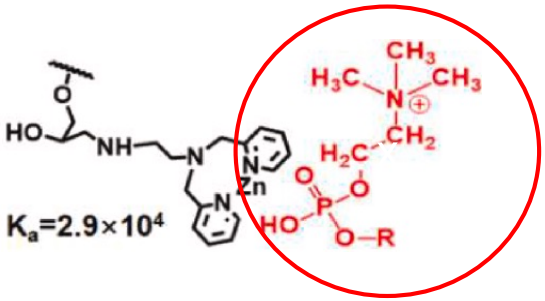


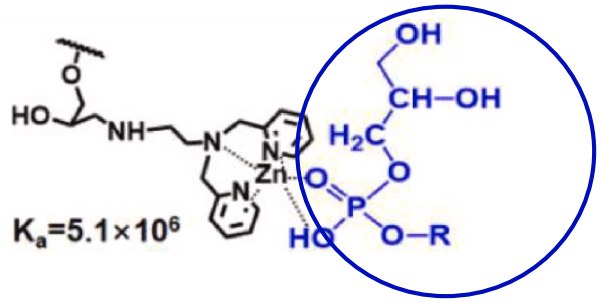
A dipicolyl-derivative of vancomycin, Dipi-van, which can chelate Zn^{2+} resulting in the formation of Dipi-van–Zn^{2+} complexes with the pyrophosphates of cell-wall lipids.

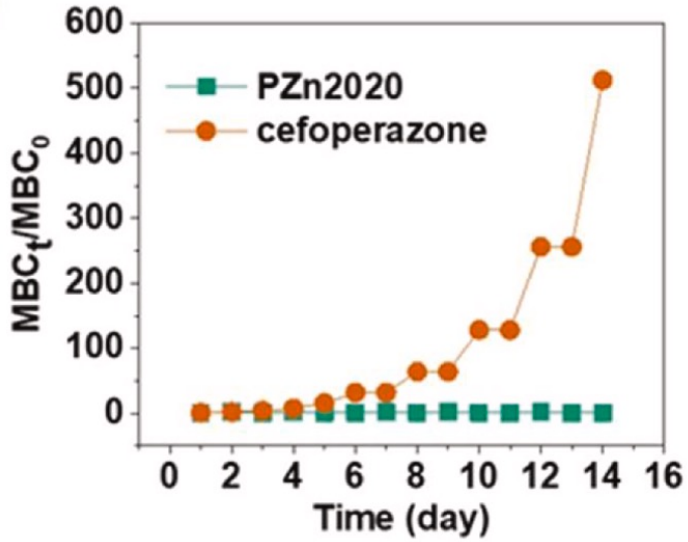
(Angew Chem Int Ed Engl, 2016)

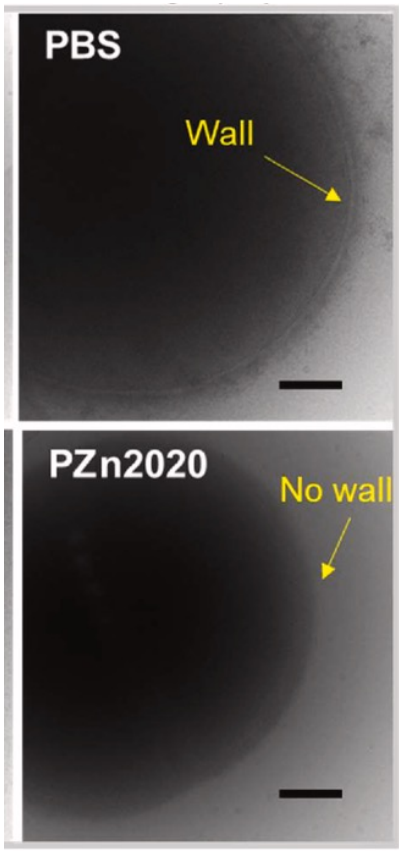
Resistance evasive antibiotics

PPi-caging PZn2020







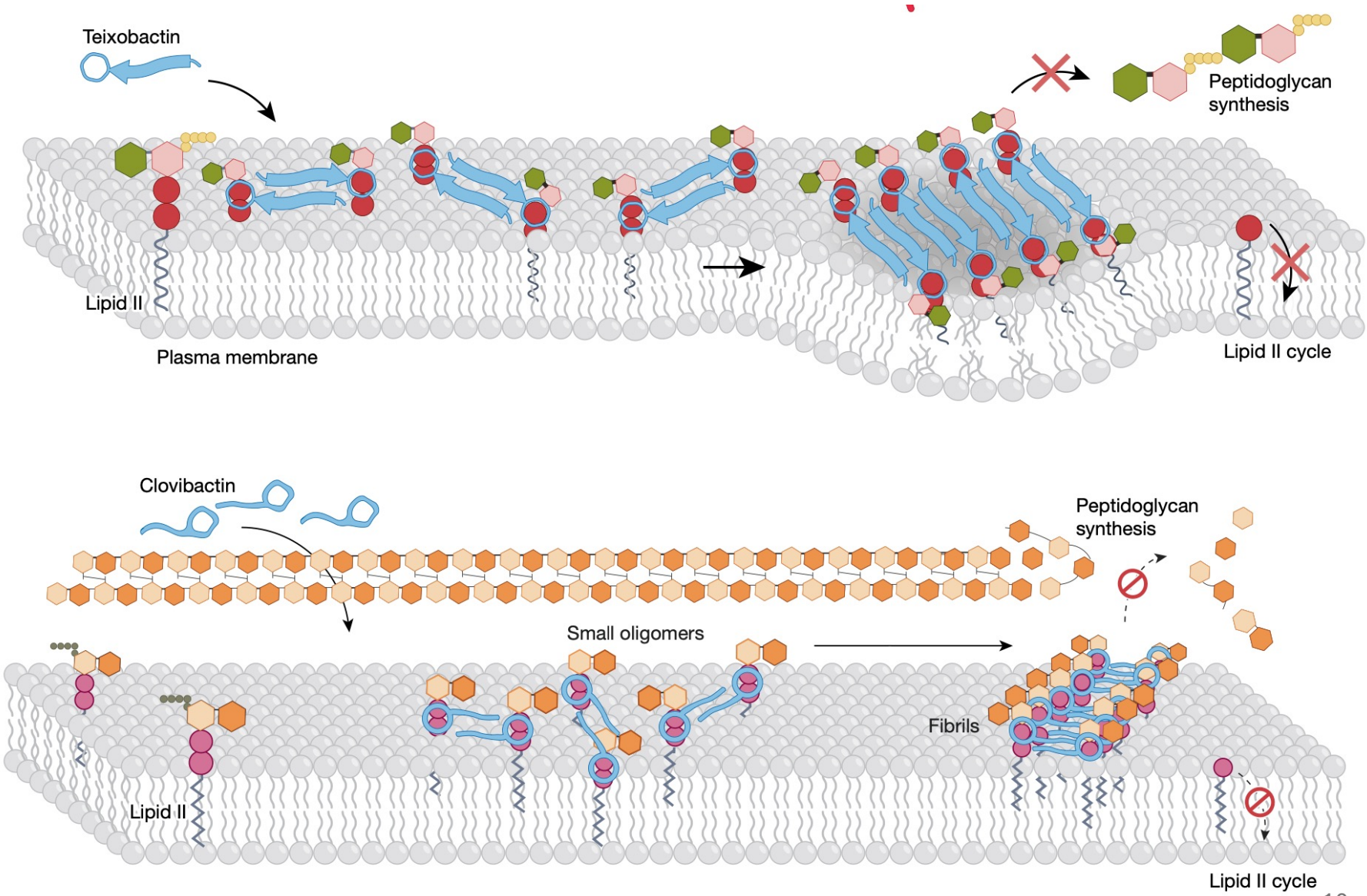


- Neutral phospholipids, phosphatidylcholine
- Negatively charged phospholipids, phosphatidylglycerol;

(Chemical Engineering Journal, 2022)

Resistance evasive antibiotics

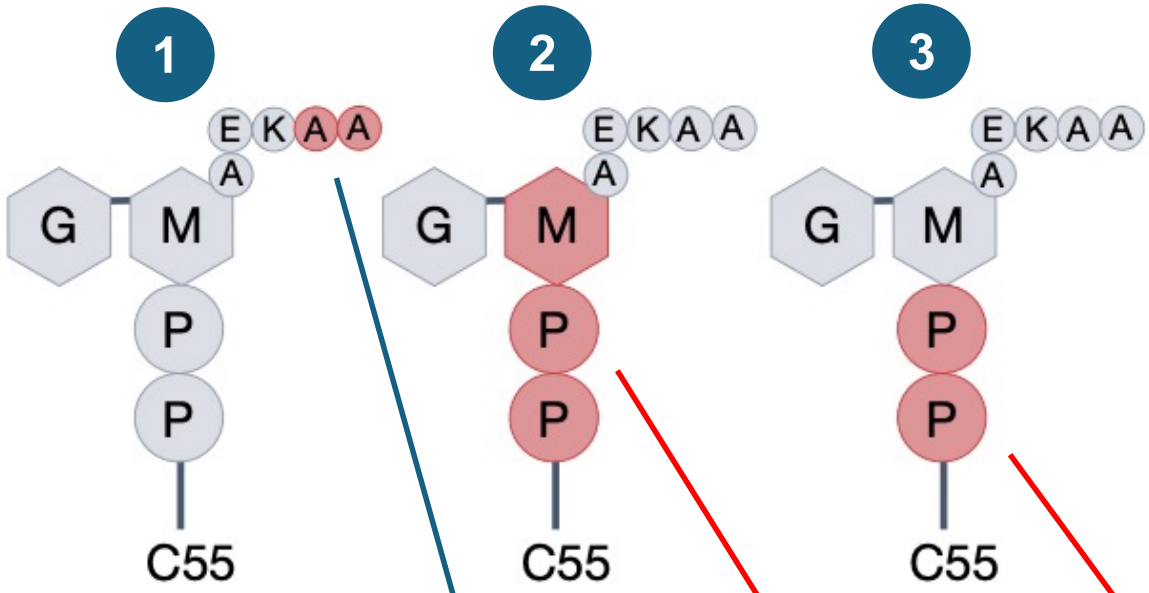
Natural antimicrobial peptides found in uncultivable bacteria.



(Nature, 2015; Cell, 2023; Nature 2024)

Resistance evasive antibiotics

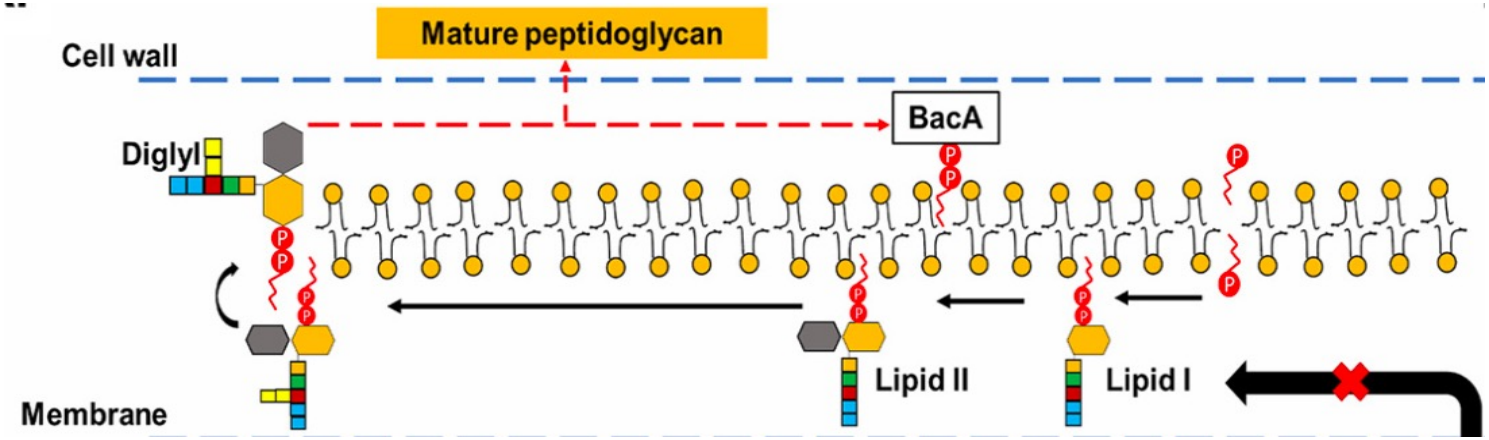
- **Vancomycin**
- **Teixobactin**
- **Clovibactin**

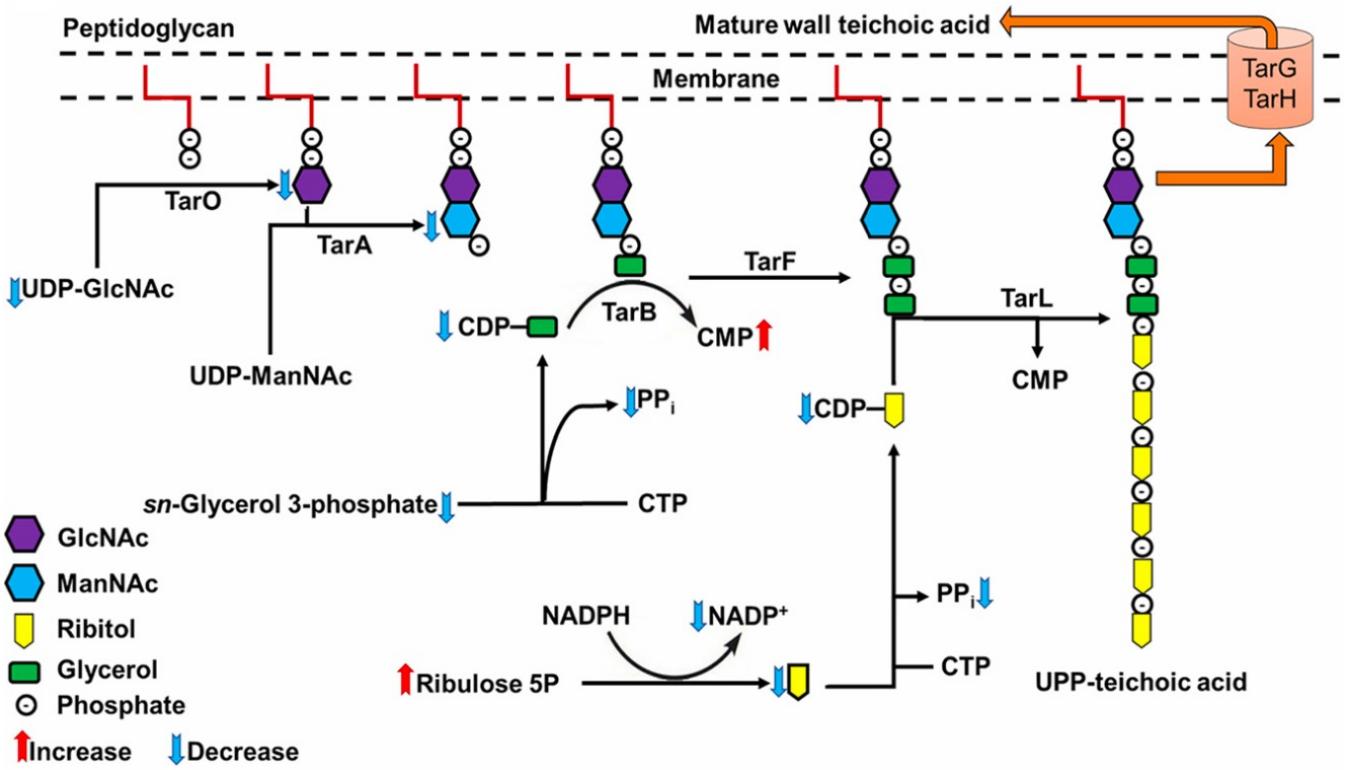


- Lipid II is immutable, but the pentapeptide is modifiable;
- MurNAc sugar moiety is conserved, but it is conceivably modifiable;
- Pyrophosphate is the end of the road for the evolution of resistance evasion: present in all lipid II-type compounds, completely immutable target;

(Nature, 2015; Cell, 2023; Nature 2024)

PPi participated cell wall biosynthesis





(mSystems, 2020)

Induction of ROS: A strategy for the control of ARG dissemination

Horizontal gene transfer, acquisition of foreign DNA material through HGT is one of the most important drivers of bacterial evolution and it is frequently responsible for the development of antimicrobial resistance.

UV/H_2O_2, ozonation, photocatalysis, and Fenton reaction have been successfully used to kill antibiotic resistant bacteria, and more importantly repress the dissemination of **antibiotic resistant genes**.

ROS refers to reactive radicals, including superoxide anion(O_2−), peroxide(O_2^{2-}), hydrogen peroxide (H_2O_2), hydroxyl radicals (OH•), and hydroxyl ions(OH^-) that are produced continually as alternative metabolites of several cell biological pathways.

Lipid autoxidation:

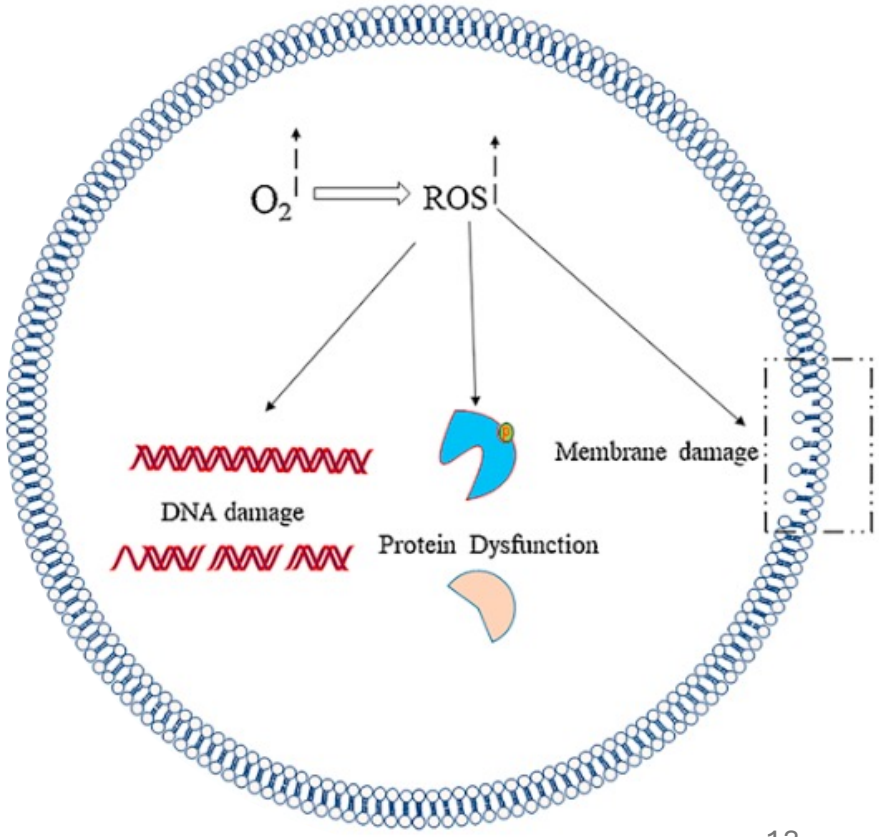
R_2N_2 is azo compound; LH is lipid substrate;

Initiation:
$R_2N_2 \rightarrow 2R\cdot + N_2$
$R\cdot + O_2 \rightarrow ROO\cdot$
$ROO\cdot + LH \rightarrow ROOH + L\cdot$

Propogation:
$L\cdot + O_2 \rightarrow LOO\cdot$
$LOO\cdot + LH \rightarrow LOOH + L\cdot$

Inhibition:
$LOO\cdot + LOO\cdot \rightarrow$ nonradical products

Fenton reaction: $H_2O_2 + Fe^{2+} \rightarrow HO\cdot + Fe^{3+} + OH^-$

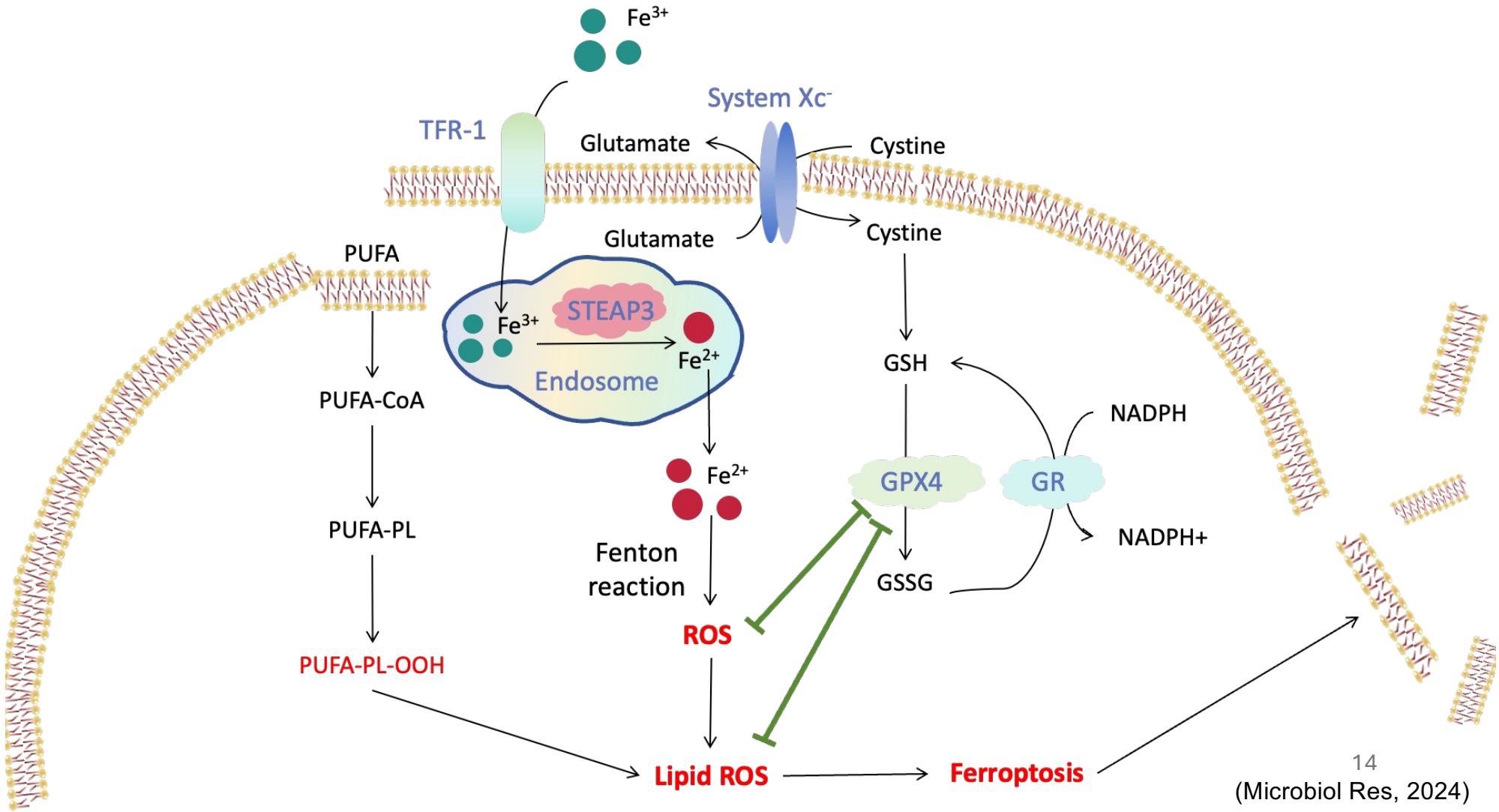


(Biomed Pharmacother, 2019)

Ferroptosis/Cuproptosis-like bacterial cell death

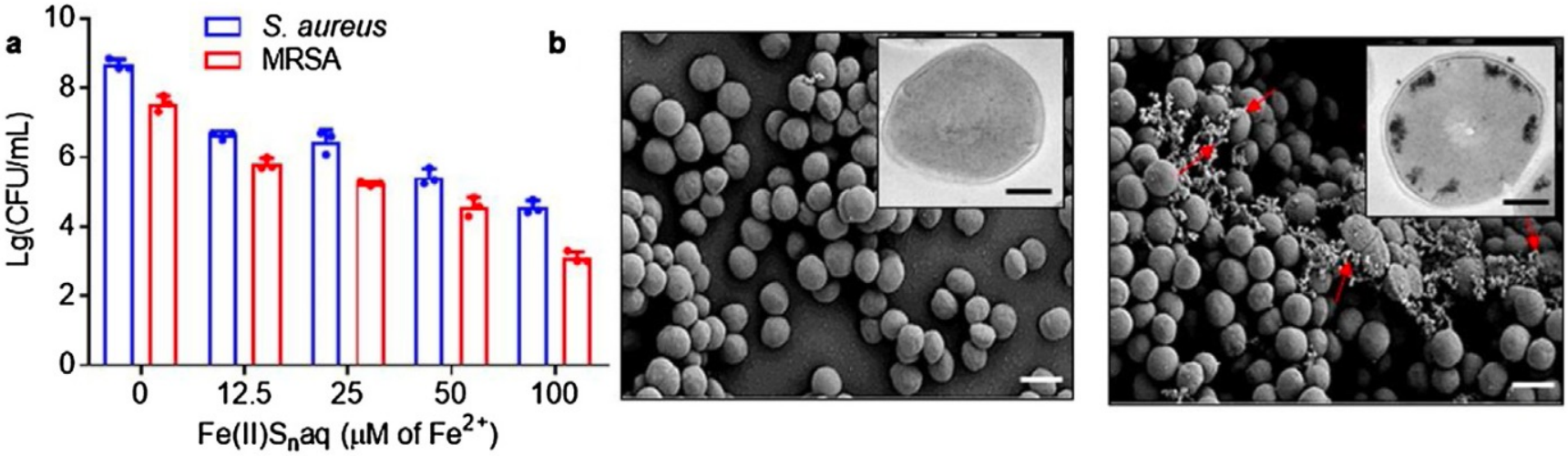
Irons and ROS generated from lipid and iron metabolism are the two key substances involved in ferroptosis.

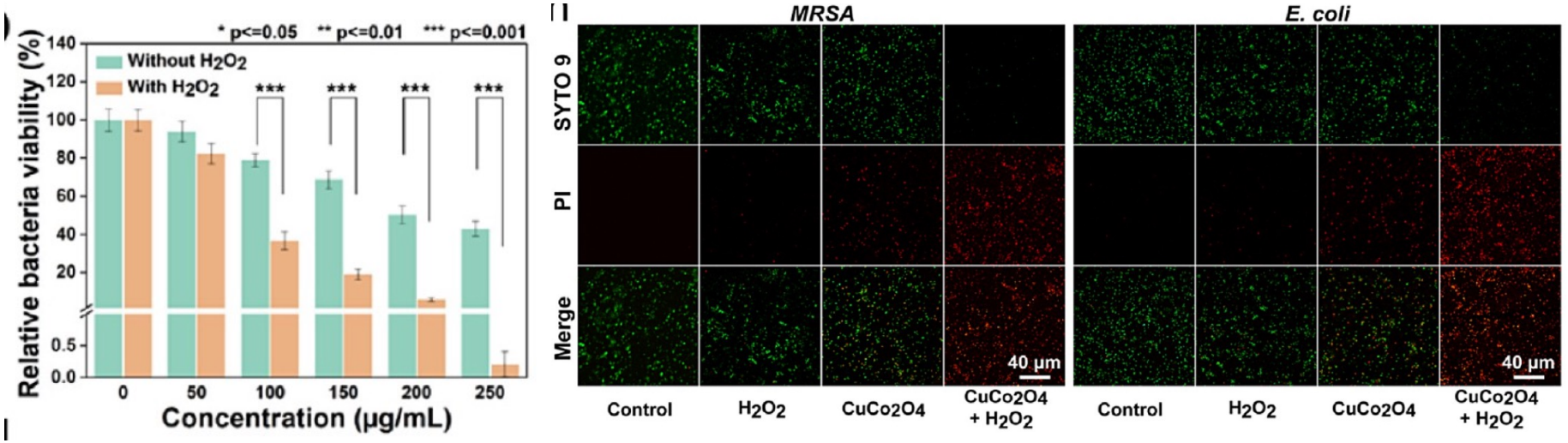
Iron catalyzes the formation of highly reactive hydroxyl radical through Fenton reaction. It further acts as a cofactor for lipoxygenase to catalyze PUFA oxidation, exerting oxidative damage in cells



(Microbiol Res, 2024)

Ferroptosis/Cuproptosis-like bacterial cell death

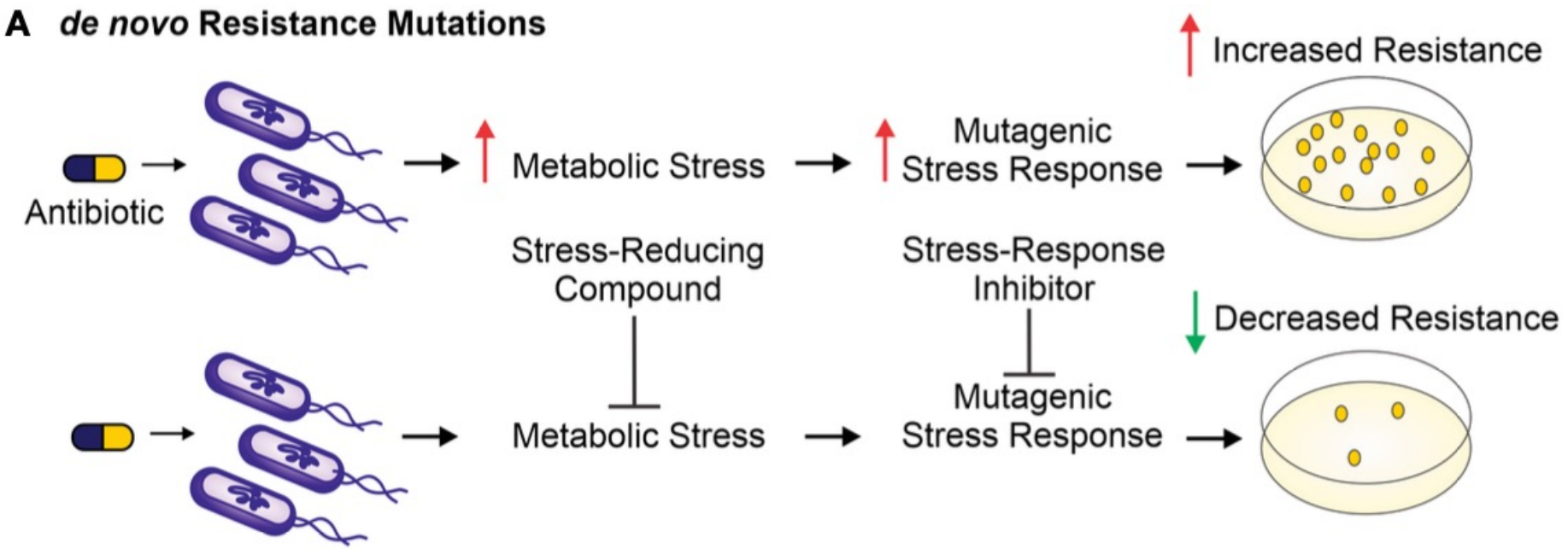




(ACS Nano, 2024; Microbiol Res, 2024; Materials & Design, 2024)

Drawbacks and Conclusion

- Clovibactin as a first-generation antibiotic may have the potential for long-term effectiveness, it is imperative to exercise caution, maintain continual monitoring, and undertake further research to gain a comprehensive understanding of the potential pathways of resistance development.



- The DNA repair process can involve the activation of mutagenic DNA polymerases that lack proofreading activity potentially leading to resistance-conferring mutations

References

1. Yarlagadda V, Sarkar P, Samaddar S, Haldar J. A Vancomycin Derivative with a Pyrophosphate-Binding Group: A Strategy to Combat Vancomycin-Resistant Bacteria. Angew Chem Int Ed Engl 2016; 55(27): 7836-40.
2. Wang W, Cui Y, Wei X, et al. CuCo(2)O(4) Nanoflowers with Multiple Enzyme Activities for Treating Bacterium-Infected Wounds via Cuproptosis-like Death. ACS Nano 2024; 18(24): 15845-63.
3. Wang H, Guo J, Yang Y, et al. CuFeS2 nanozyme regulating ROS/GSH redox induces ferroptosis-like death in bacteria for robust anti-infection therapy. Materials & Design 2024; 239.
4. Toussaint KA, Gallagher JC. beta-lactam/beta-lactamase inhibitor combinations: from then to now. Ann Pharmacother 2015; 49(1): 86-98.
5. Sun S, Chen X. Mechanism-guided strategies for combating antibiotic resistance. World J Microbiol Biotechnol 2024; 40(10): 295.
6. Stelitano G, Cocorullo M, Mori M, Villa S, Meneghetti F, Chiarelli LR. Iron Acquisition and Metabolism as a Promising Target for Antimicrobials (Bottlenecks and Opportunities): Where Do We Stand? Int J Mol Sci 2023; 24(7).
7. Shukla R, Medeiros-Silva J, Parmar A, et al. Mode of action of teixobactins in cellular membranes. Nat Commun 2020; 11(1): 2848.
8. Shen X, Ma R, Huang Y, et al. Nano-decocted ferrous polysulfide coordinates ferroptosis-like death in bacteria for anti-infection therapy. Nano Today 2020; 35.
9. Oldfield E, Feng X. Resistance-resistant antibiotics. Trends Pharmacol Sci 2014; 35(12): 664-74.
10. Nie X, Gao F, You W, et al. Caging pyrophosphate structure blocks the cell wall synthesis to kill bacteria without detectable resistance. Chemical Engineering Journal 2022; 450.
11. Munita JM, Arias CA. Mechanisms of Antibiotic Resistance. Microbiol Spectr 2016; 4(2).
12. Memar MY, Yekani M, Alizadeh N, Baghi HB. Hyperbaric oxygen therapy: Antimicrobial mechanisms and clinical application for infections. Biomed Pharmacother 2019; 109: 440-7.
13. Luo Z, Lu R, Shi T, et al. Enhanced Bacterial Cuproptosis-Like Death via Reversal of Hypoxia Microenvironment for Biofilm Infection Treatment. Adv Sci (Weinh) 2024; 11(19): e2308850.
14. Lewis K, Lee RE, Brotz-Oesterhelt H, et al. Sophisticated natural products as antibiotics. Nature 2024; 632(8023): 39-49.
15. Lewis K. At the Crossroads of Bioenergetics and Antibiotic Discovery. Biochemistry (Mosc) 2020; 85(12): 1469-83.
16. Jekhmane S, Derks MGN, Maity S, et al. Host defence peptide plectasin targets bacterial cell wall precursor lipid II by a calcium-sensitive supramolecular mechanism. Nat Microbiol 2024; 9(7): 1778-91.
17. Hsu ST, Breukink E, Tischenko E, et al. The nisin-lipid II complex reveals a pyrophosphate cage that provides a blueprint for novel antibiotics. Nat Struct Mol Biol 2004; 11(10): 963-7.
18. Guo R, Fang X, Shang K, Wen J, Ding K. Induction of ferroptosis: A new strategy for the control of bacterial infections. Microbiol Res 2024; 284: 127728.
19. Darby EM, Trampari E, Siasat P, et al. Molecular mechanisms of antibiotic resistance revisited. Nat Rev Microbiol 2023; 21(5): 280-95.
20. Cheng Z, He BB, Lei K, et al. Rule-based omics mining reveals antimicrobial macrocyclic peptides against drug-resistant clinical isolates. Nat Commun 2024; 15(1): 4901.
21. Butler MS, Vollmer W, Goodall ECA, Capon RJ, Henderson IR, Blaskovich MAT. A Review of Antibacterial Candidates with New Modes of Action. ACS Infect Dis 2024; 10(10): 3440-74.
22. Batchelder JI, Hare PJ, Mok WWK. Resistance-resistant antibacterial treatment strategies. Front Antibiot 2023; 2.

Q&A