

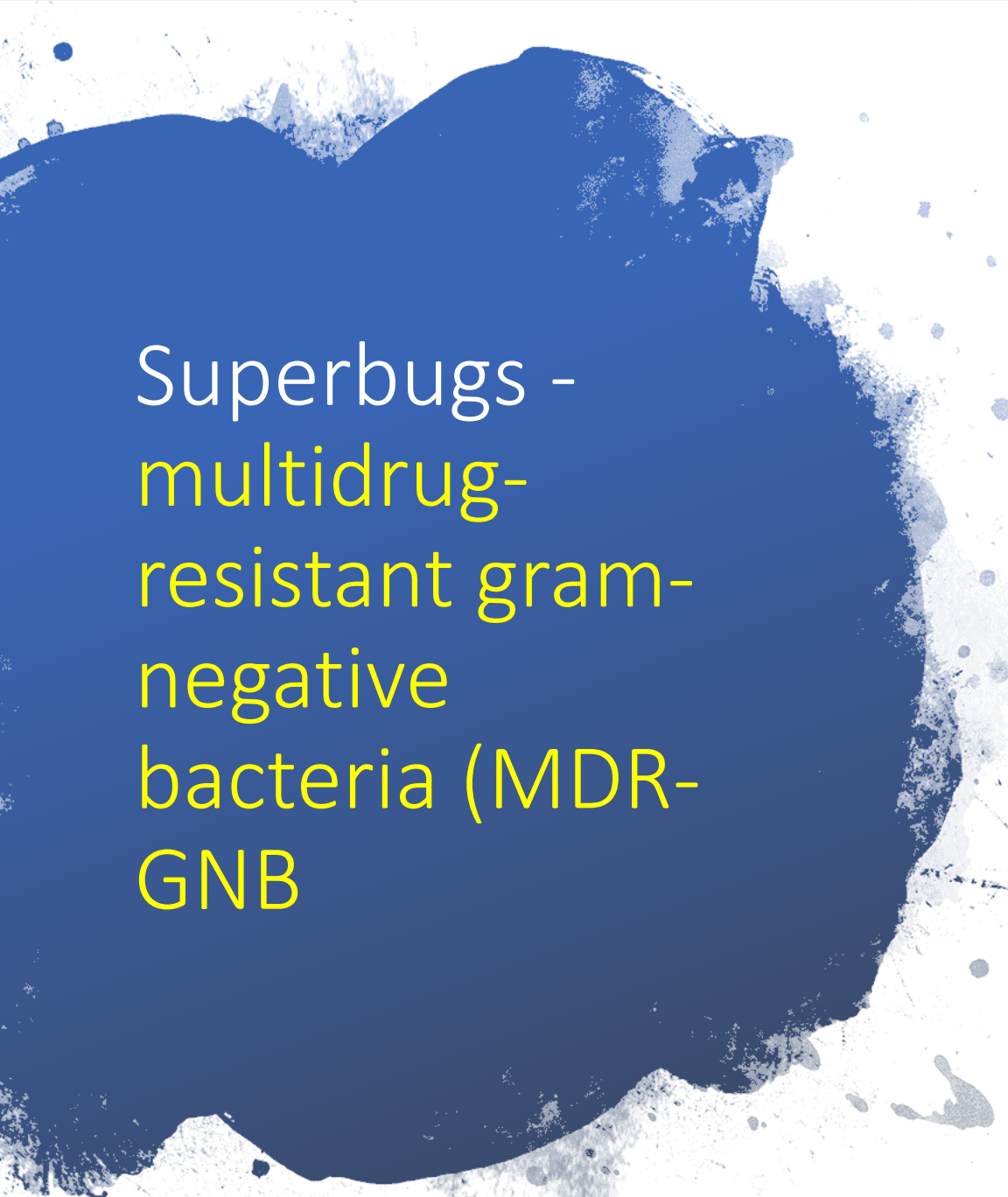
MULTISCALE  
MODELING AND  
ANALYSIS OF  
LIPID  
MEMBRANES  
INTERACTION  
WITH  
ANTIBIOTICS : A  
CASE STUDY ON  
GRAM-NEGATIVE  
BACTERIA

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- The Swedish National Infrastructure for Computing (SNIC) , Sweden
- National Supercomputer Centre at Linköping University, Sweden
- European Structural Funds, Government of the Republic of Lithuania

- Antibiotic resistance kills 700,000 people every year:  
HORIZON: the EU Research & Innovation magazine

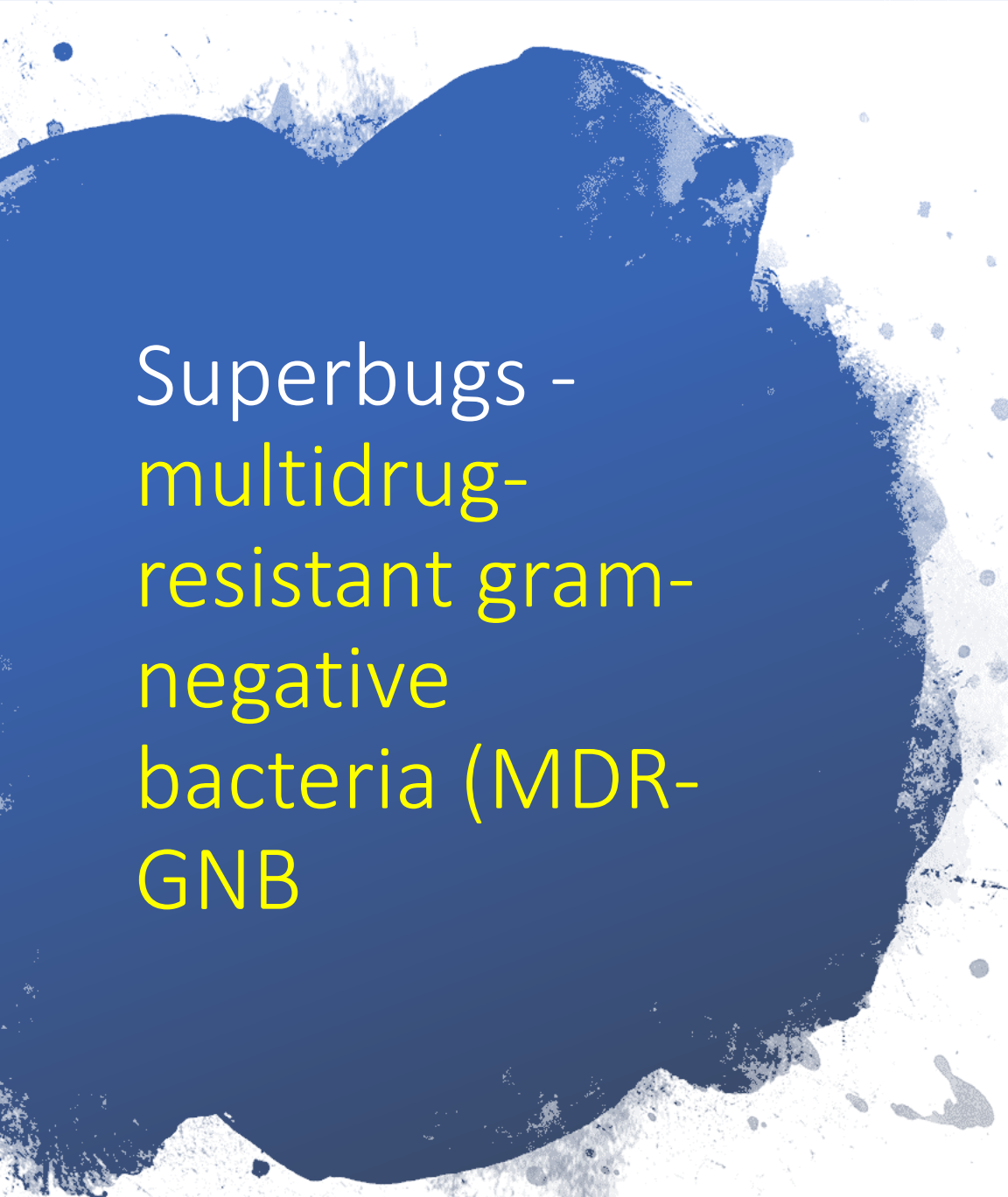
- Antimicrobial resistance could kill 10 million people every year by 2050: HORIZON: the EU Research & Innovation magazine

- Preliminary results generated using computational methods – molecular dynamics simulations



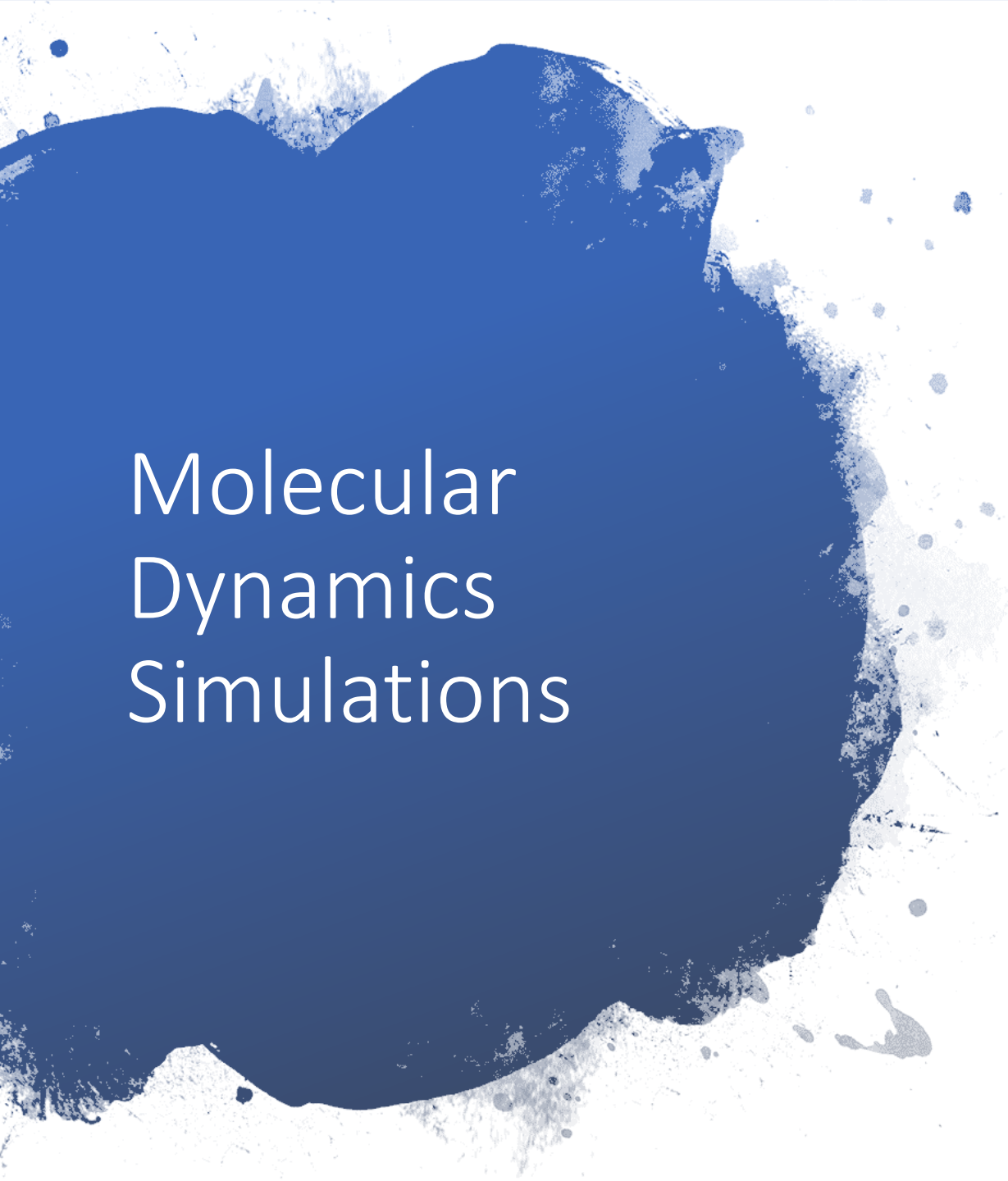
# Superbugs - multidrug- resistant gram- negative bacteria (MDR- GNB

- Gram-negative bacteria, the **superbugs** are already attaining resistance to broad range of antibiotics leading to chronic infections and increase in the cost of healthcare worldwide and biofilm associated infections.
- Various diseases are caused due to membrane abnormalities. It is proved that **biological membranes are good targets for therapy.**(1).
- The bacterial interaction with their surroundings are decided through the membranes that is a part of the cell envelopes. These envelopes are highly complex structures though bacteria can be simple organisms (2).



# Superbugs - multidrug- resistant gram- negative bacteria (MDR- GNB)

- Further in patients with cancer, blood stream infections are responsible for major complications due to the delays in chemotherapy along with many others. **Gram-negative bacteria** is associated with the etiology of BSI (3).
- Studies have been conducted evaluating the predictors and outcomes associated with multidrug-resistant gram-negative bacterial (MDR-GNB) infections in an oncology pediatric intensive care unit (PICU).
- Treatment of infections caused by multidrug-resistant Gram-negative bacteria: report of the British Society for Antimicrobial Chemotherapy/Healthcare Infection Society/British Infection Association Joint Working Party.



# Molecular Dynamics Simulations

- **Molecular dynamics (MD)** simulations help in deriving structural and dynamic descriptions of membrane than it is possible by conducting experiments. Even though it is highly essential to validate the results in agreement with the available experimental data.
- **Steered molecular dynamics (SMD)** induces unbinding of ligands and conformational changes in biomolecules on time scales accessible to **molecular dynamics simulations**. Time-dependent external forces are applied to a system, and the responses of the system are analyzed.





# Nanoparticles

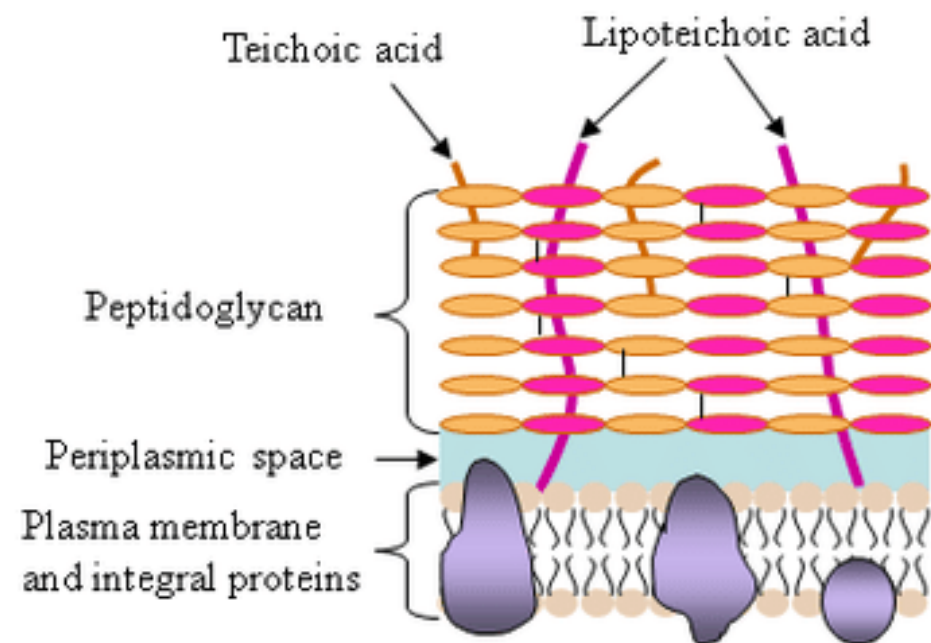
- Nanoparticles as such are being used as carrier for transporting the antibiotics to a specific target site to deliver the drugs or compounds used against gram-negative bacteria. Recent studies have proved the use of nanomaterials as a technique that can be adopted for drug delivery systems.



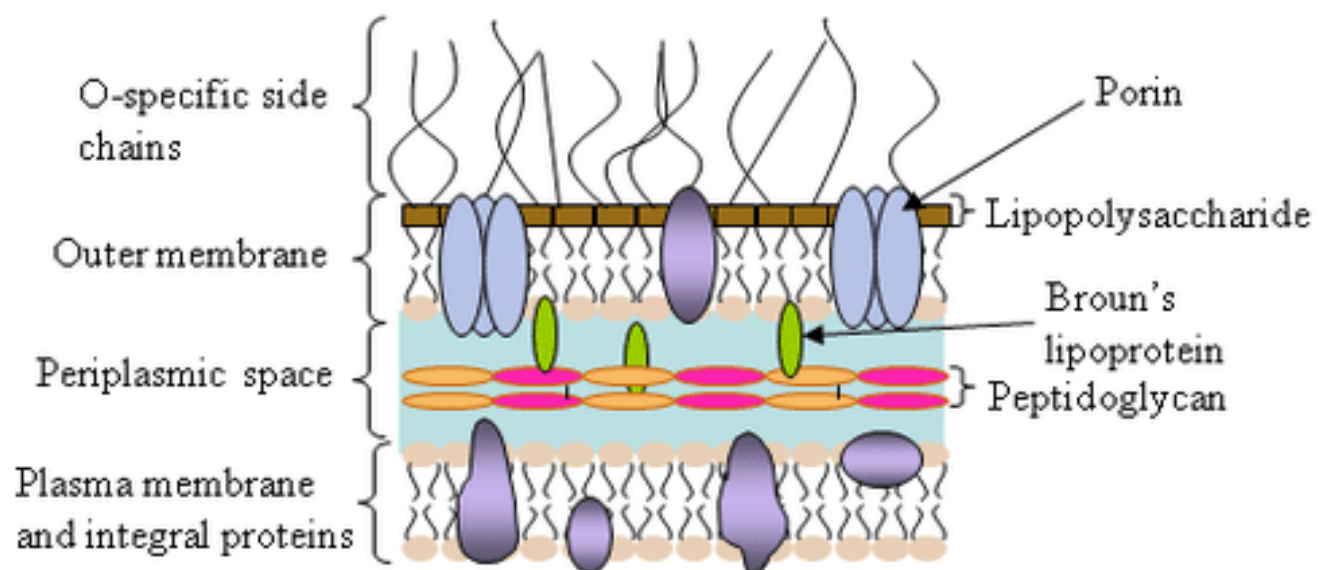
# Development of a predictive model for antimicrobial activity of titanium dioxide structures surfaces

- “Despite numerous existing potent antibiotics and other antimicrobial means, **bacterial infections are still a major cause of morbidity, implant rejection, and mortality.**
- Moreover, the need to develop additional bactericidal means has significantly increased due to the growing concern regarding multidrug-resistant bacterial strains and biofilm associated infections.
- **Bacterial adhesion onto solid surfaces is the onset of biofilm formation**, which is of critical importance in a wide spectrum of problems including many modern industrial applications, especially in microbiology and biomedical research.
- The mechanism of bacterial adhesion to solid surfaces is a complex process affected by multiple factors. The most important property of bacterial adhesion mechanism is influenced by **physicochemical interaction** of the bacterial cells.
- The microscopic understanding of bacteria cell envelope interaction with its surroundings is one of the corner stones of **rational design of novel antibiotics and antimicrobial coatings.**
- Unfortunately, the complexity of bacterial cell envelopes prevented until now **development of multiscale models for these systems, and only recently progress had been made in coarse grain molecular dynamic simulations of cell envelopes of gram-negative bacteria”**
- - Prof. Rinkevicius, Prof. Laukaitis - **Project no.:** 09.3.3-LMT-K-712-01-0162

### Gram (+) cell-wall



### Gram (-) cell-wall



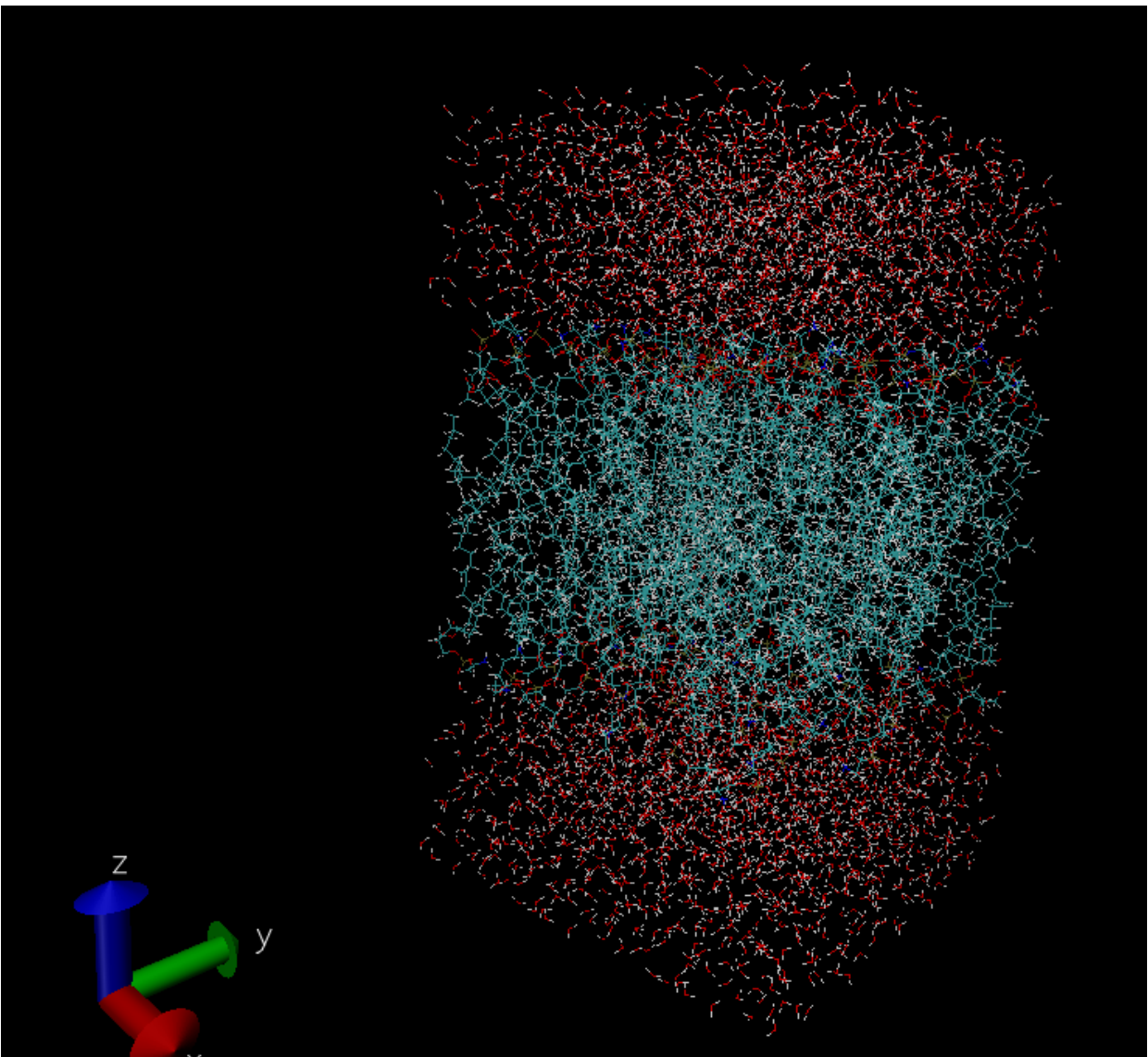
# Introduction

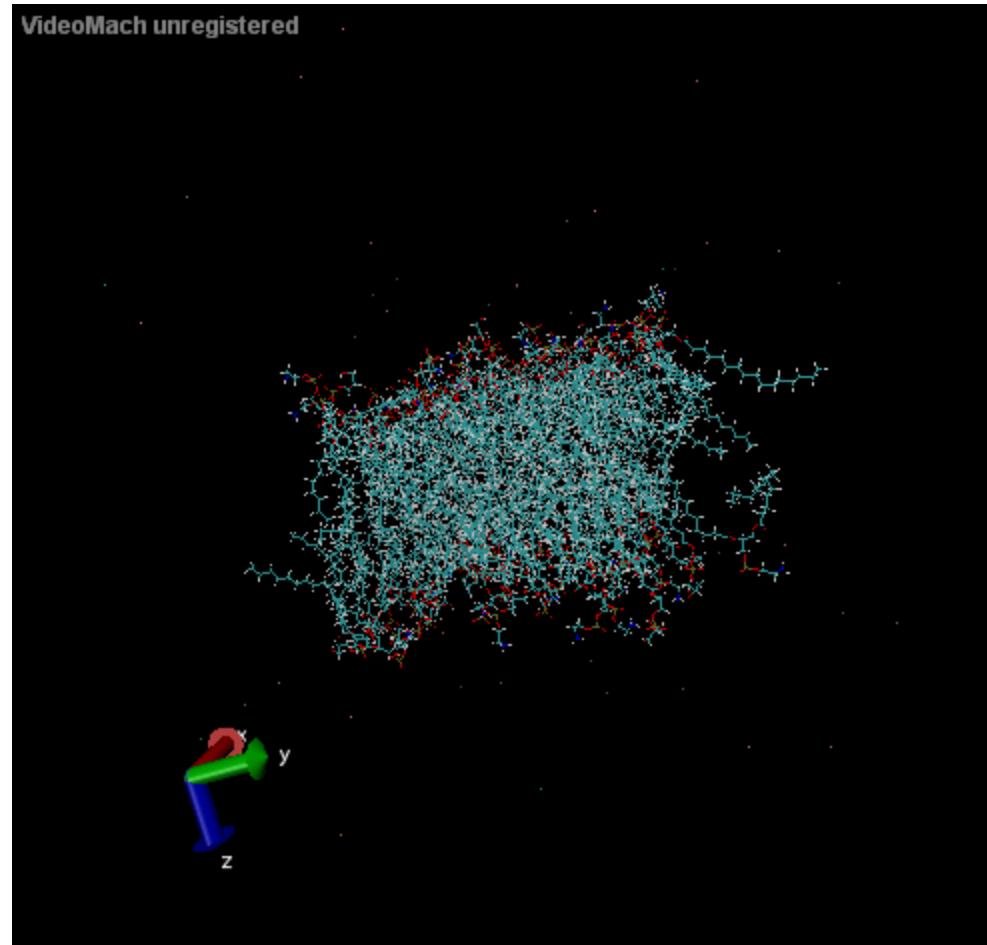
- Lipids are a significant component of lipid bilayers. They play significant role in many **cell signaling and physiological processes**.
- “Lipopolysaccharides (LPS) are an integral component of the outer leaflet of the **outer membrane of Gram-negative bacteria** where they function/act as a shield and barrier to environmental threats to the bacterium.
- The study of LPS and the overall outer membrane shape and structure is important because LPS structure can inform outer **membrane protein biogenesis and antibiotic design**.
- The shape and stability of the outer membrane is believed to play a large part in the insertion and folding of outer membrane proteins.
- It is the LPS that make the outer membrane more **impermeable to antibiotics**, so understanding and then manipulating LPS membrane stability would be a **rational way to develop new Gram-negative antibiotics**.”

Reference: S. Kim, D.S. Patel, S. Park, J. Slusky, J.B. Klauda, G. Widmalm, W. Im. **Bilayer properties of lipid a from various gram-negative bacteria**. Biophys J, 111 (2016), pp. 1750-1760

Calculated Number of Lipids: 1:1		
Lipid Type	Upperleaflet Number	Lowerleaflet Number
POPE	21	21
POPG	21	21

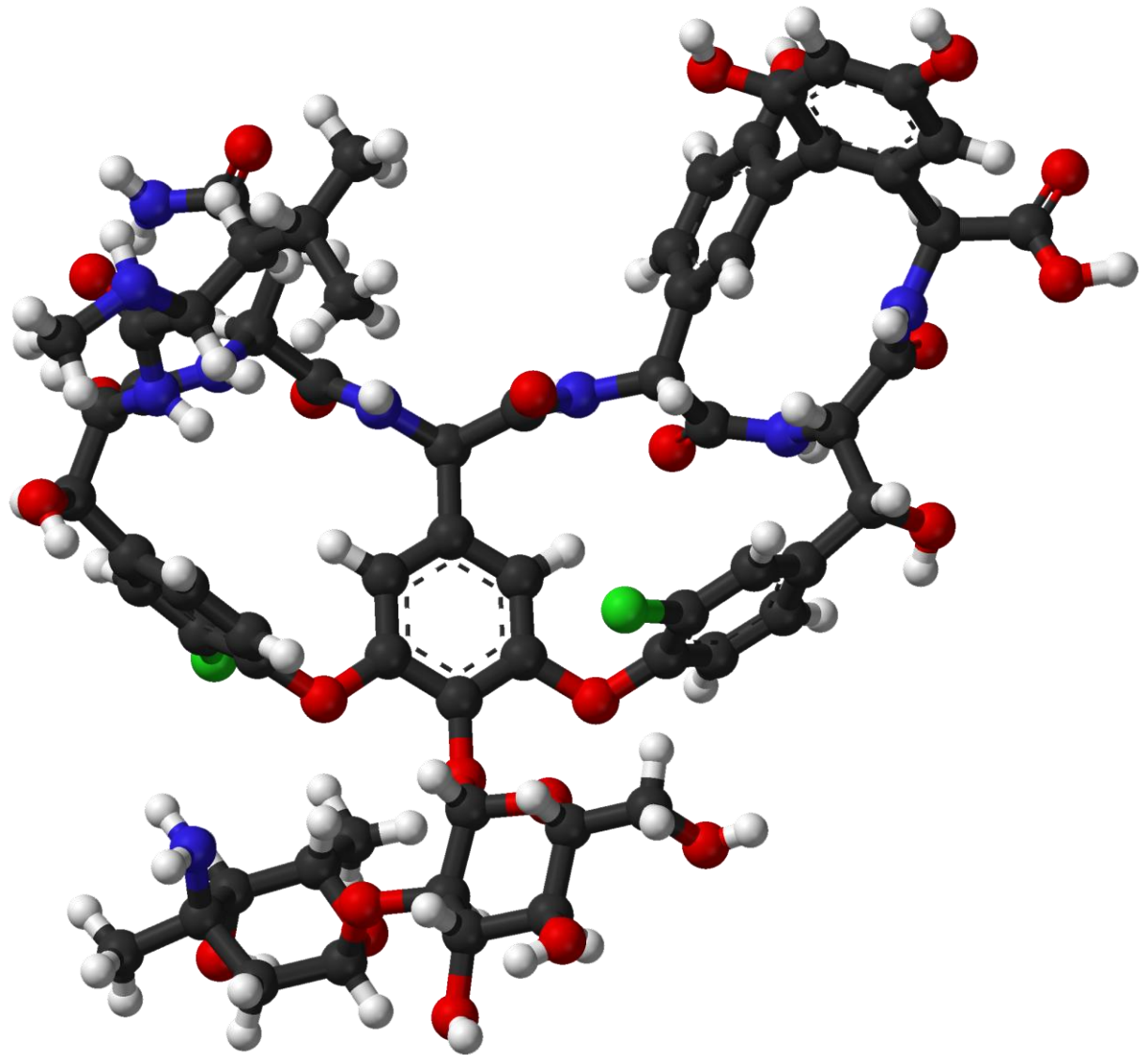
- Calculated XY System Size:
- |              |              |              |
|--------------|--------------|--------------|
|              | Upperleaflet | Lowerleaflet |
| Protein Area | 0            | 0            |
| Lipid Area   | 2536.8       | 2536.8       |
| # of Lipids  | 42           | 42           |
| Total Area   | 2536.8       | 2536.8       |
| Average Area | 2536.80      | 2536.80      |
| A            | 50.37        | 50.37        |
| B            | 50.37        | 50.37        |





# Vancomycin

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# Vancomycin

- Vancomycin is an antibiotic used to treat many bacterial infections
- It is used against Gram-positive infections unresponsive to other antibiotics
- Till date usage of vancomycin against Gram-negative infections is not well documented
- The mechanism by which Gram-negative bacteria produce their cell wall is different along with other factors related to the entering of outer membranes



# Nanotransformation

- *Fernandes et al., ACS Appl. Mater. Interfaces 2017, 9, 17, 15022-15030*

- Nanotechnology has been considered as a novel tool for overcoming the development of microbial resistance
- Antibacterials processed into nanosized particles are claimed to be more efficient than their nonprocessed counterparts
- The exact mechanisms by which nanomaterials interact with bacteria are not fully understood, but it is thought that the nanoparticles cause an increase in the microbial cell membrane permeability, osmotic damage, and flow of cytoplasmic components out of the cell, eventually leading to cell death

# Nanotransformation

- *Fernandes et al., ACS Appl. Mater. Interfaces* 2017, 9, 17, 15022-15030

- Nanospherization extended the effect of vancomycin toward Gram-negative *Escherichia coli* and *Pseudomonas aeruginosa*, making these bacteria up to 10 and 100 times more sensitive to the antibiotic, respectively.
- The spheres were able to disrupt the outer membranes of these bacteria, overcoming their intrinsic resistance toward glycopeptides.
- The penetration of nanospheres into a Langmuir monolayer of bacterial membrane phospholipids confirmed the interaction of the nanoantibiotic with the membrane of *E. coli* cells, affecting their physical integrity, as further visualized by scanning electron microscopy
- The nanotransformation of vancomycin could overcome the inherent resistance of Gram-negative bacteria toward this antibiotic and disrupt mature biofilms at antibacterial-effective concentrations.



# Analysis

- Structure of membrane intact
- Vancomycin movement to be monitored
- Structure of the ligand is intact ?
- Larger structure of the ligand – difficult to penetrate
- Tag with fluorophore to track the movement
- Steered dynamics

# Results and perspectives

We assessed the sensitivity of the structure and dynamics of the bilayer using molecular dynamics simulations using multiple parameters and by choosing a Amber force field

We realized the structure is stable after MD simulations and the chosen force field is appropriate and did not disintegrate the system

Bilayer model can accommodate organic small molecules that will allow the entry of the molecules into the system

It is also possible  $\text{TiO}_2/\text{Au}$  nanoparticles could be introduced into the system

## Thank you

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