Computational Investigation of Claudin-3 and the effects of Palmitoylation

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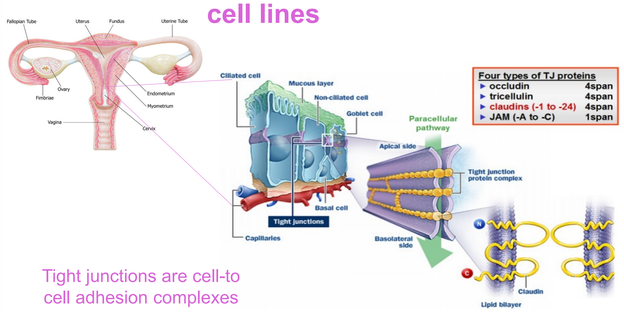
Introduction

- Claudin-3 is a membrane protein that plays a critical role in maintaining size and charge selectivity in tight junctions

Claudin-3 is expressed in a panel of ovarian tumors of various subtypes and cell lines



Tight junctions are cell-to cell adhesion complexes

Claudin-3 (human) primary sequence

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        10         20         30         40         50
MSMGLEITGT ALAVLGWLGT IVCCALPMWR VSAFIGSNII TSQNIWEGLW
        60         70         80         90        100
MNCVVQSTGQ MQCKVYDSLL ALPQDLQAAR ALIVVAILLA AFGLLVALVG
       110        120        130        140        150
AQCTNCVQDD TAKAKITIVA GVLFLLAALL TLVPVSWSAN TIIRDFYNPV
       160        170        180        190        200
VPEAQKREMG AGLYVGWAAA ALQLLGGALL CCSCPPREKK YTATKVVYSA
       210        220
PRSTGPGASL GTGYDRKDYV
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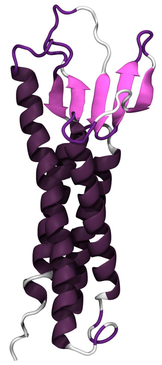
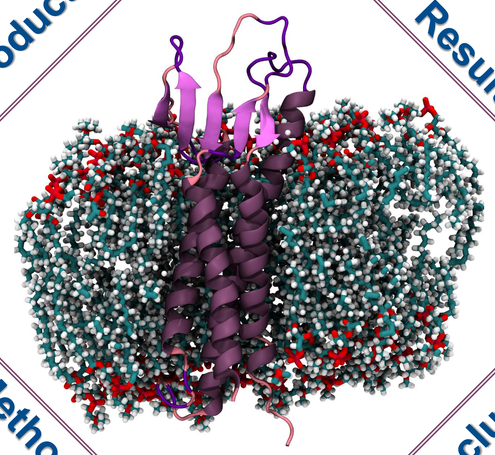
Palmitoylation sites: Cys 106, Cys 181, Cys182

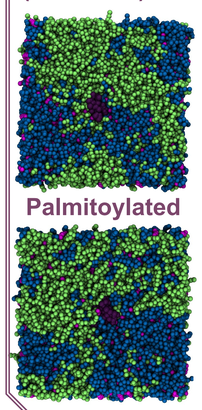


Claudin-3

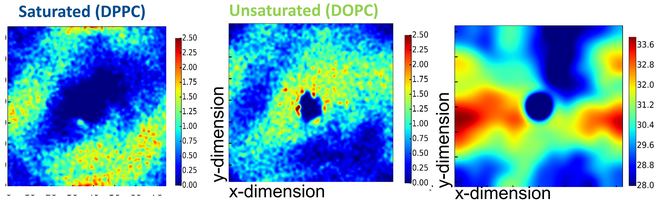


- Claudin-3 is the major component of tight junction and polymerizes to form tight junction strands with various morphologies that may correlate with their.
- Claudin-3 undergo reversible posttranslational modification at their membrane
- cysteine residues.

Results

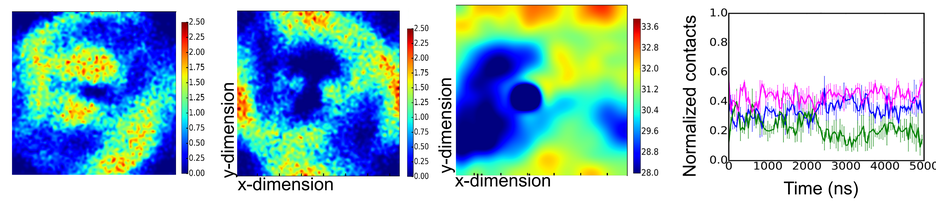


Membrane density



Membrane thickness

Lipid mixing



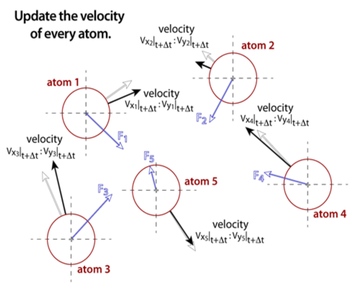
DPPC DOPC CHOL Claudin

Palmitoylation is a key posttranslational modification of Claudin-3 proteins

Method

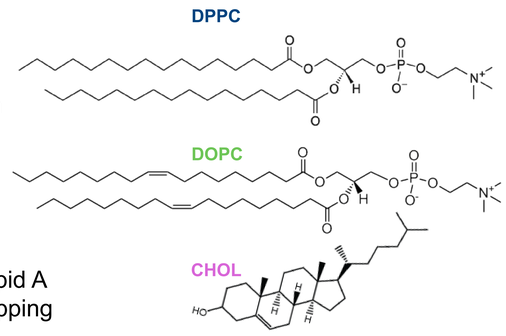
Molecular dynamics

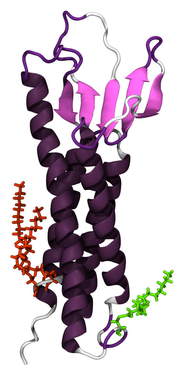
Molecular dynamics is a computational method used to simulate molecular systems.



Simulation details

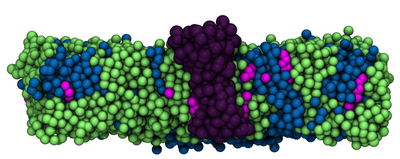
- The proteins (claudin-3 and claudin-3P) were inserted individually in membrane of a mixture of unsaturated lipid (DOPC), saturated lipid (DPPC) and cholesterol(CHOL) in a 2:2:1 ratio.
- The simulations were performed in triplicated for 10 μs each
- The coarse-grain parameterization of the library of eight lipid A molecules was developed on the Martini many-to-one mapping approach.



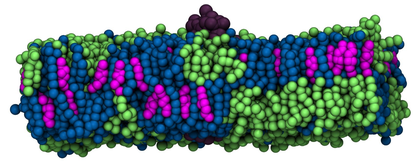


Palmitoylated Claudin-3

Conclusions



Claudin-3



Palmitoylated Claudin-3

- **Based on the results we can conclude that :**
  - NonPalmitoylated Claudin-3 localizes with unsaturated lipid domains
  - Palmitoylated Claudin-3 localizes with saturated lipids domains
  - Degree of palmitoylation enhances Claudin-3 preference for saturated lipids
  - Membrane thickness of saturated lipid domains is higher than unsaturated lipids
  - Cholesterol localizes with saturated lipids
  - The results show that claudin-3 lipid adaptation has strong correlation with its degree of palmitoylation

By understanding the behavior of this protein, we can determine its future importance in understanding diseases like Cancer !

Acknowledgments

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References

Palmitoylation of Claudin-5 Proteins Influences Their Lipid Domain Affinity and Tight Junction Assembly at the Blood–Brain Barrier Interface N. Rajagopal, F. J. Irudayanathan, and S. Nangia. *Journal of Physical Chemistry B, 122, 983–993* (2019).