# Hydration studies on lipid membranes: Classification of water molecules in different bilayers in gel and liquid phase.

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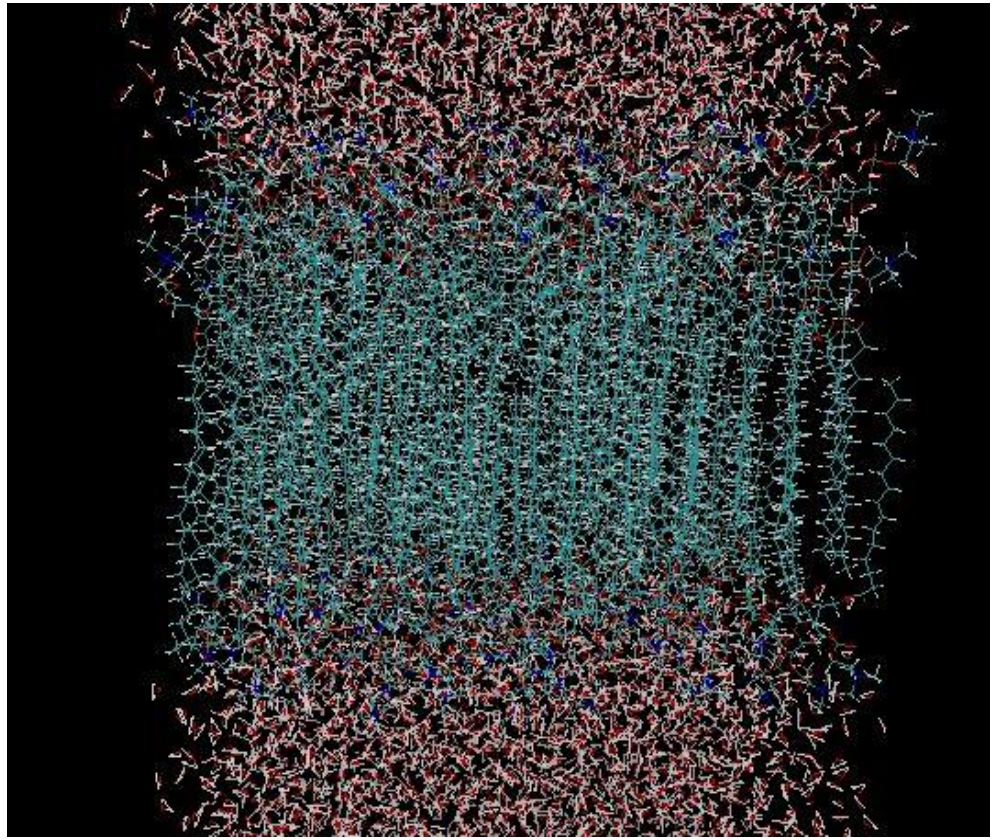
# Abstract:

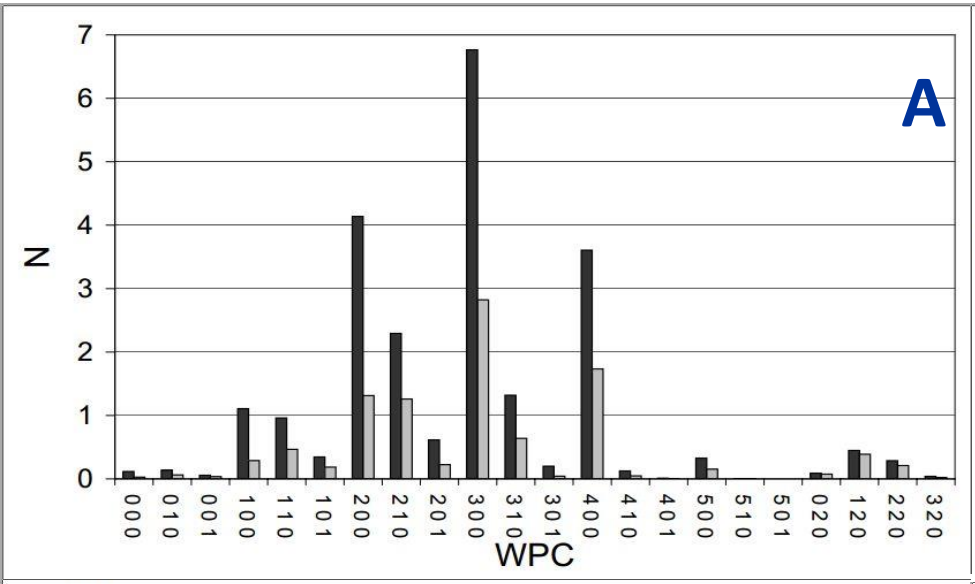
We use molecular dynamics simulations to study the hydration properties of lipid membranes, both in the gel state and in the crystalline fluid. We show that while the hydration centers remain significantly hydrated in both phases, the gel-fluid transition implies significant changes in the second hydration layer, particularly in the region of the hydrocarbon tails. Thus, while the water population is almost nil in the hydrocarbon tails zone in gel state, this region becomes partially accessible to water in crystalline liquid state in membranes of dipalmitoylphosphatidylcholine (DPPC). In the case of membranes composed of unsaturated lipids, the presence of double bonds modifies the hydration map particularly in the hydrocarbon zone. We also show that the water molecules located in the non-polar zone have a lower hydrogen bond (HB) coordination in comparison with the molecules of the first hydration layer, which are arranged in relatively compact 'nanoclusters'. The water molecules in the hydrocarbon tails zone tend to be organized in less compact structures, like branched chains, with a small population of isolated molecules. This behavior is similar to that observed in other hydration contexts, such as water penetrating carbon nanotubes or hydrophobic pores, reflecting the reluctance of water to sacrifice HB coordination.

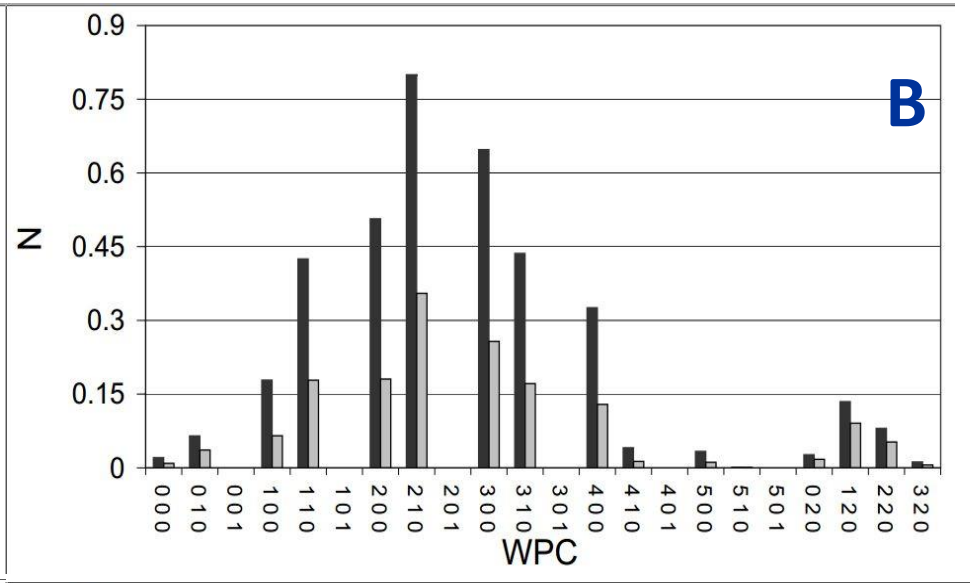
# Simulation Details:

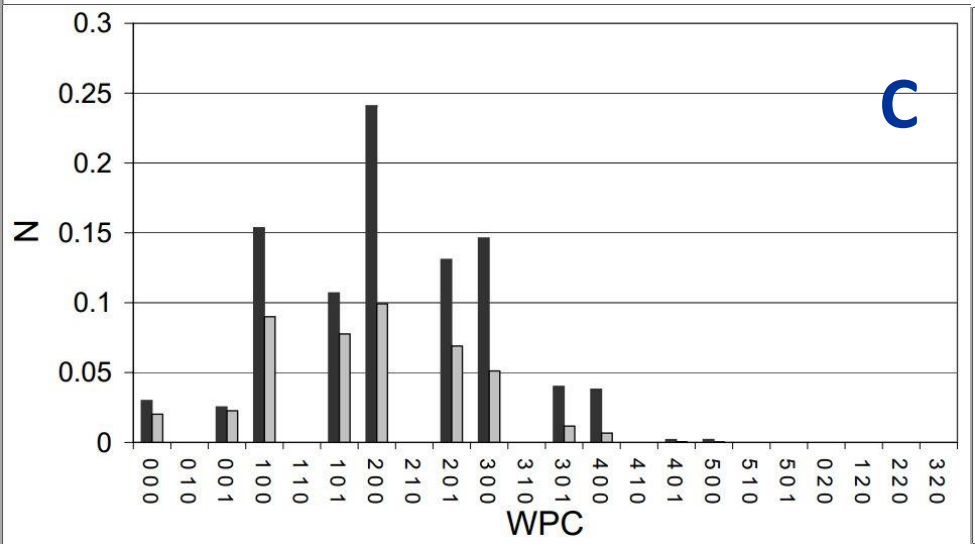
- The lipid membranes were made of 128 molecules, 64 by monolayer.
- We use dipalmitoilphosphatidilcholine (DPPC), 1-palmitoil-2-oleoilphosphatidilcholine (POPC) and dioleoilphosphatidilcholine (DOPC).
- The membranes were solvated with over a 10000 TIP3P water molecules to obtain fully hydrated systems.
- Temperature = 323 K in the fluid phase and 293 K in the gel phase.
- The number of water molecules per lipid and area per lipid were consisted with experimental data.
- Simulation were performed with the AMBER16 package with gaff2, lipid17 and water.tip3p force fields.
- The equilibrium and the final production runs were performed within the NPT ensemble.

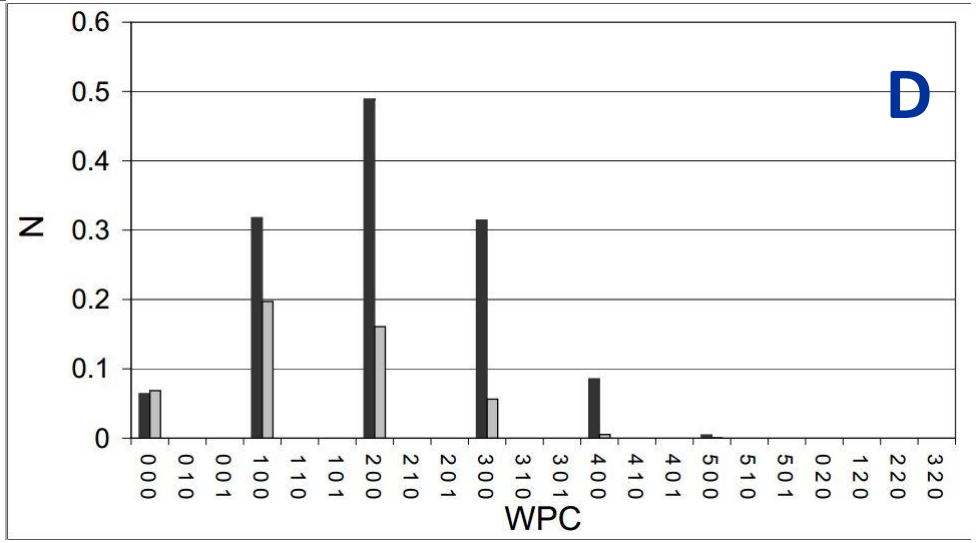


# We found different water species



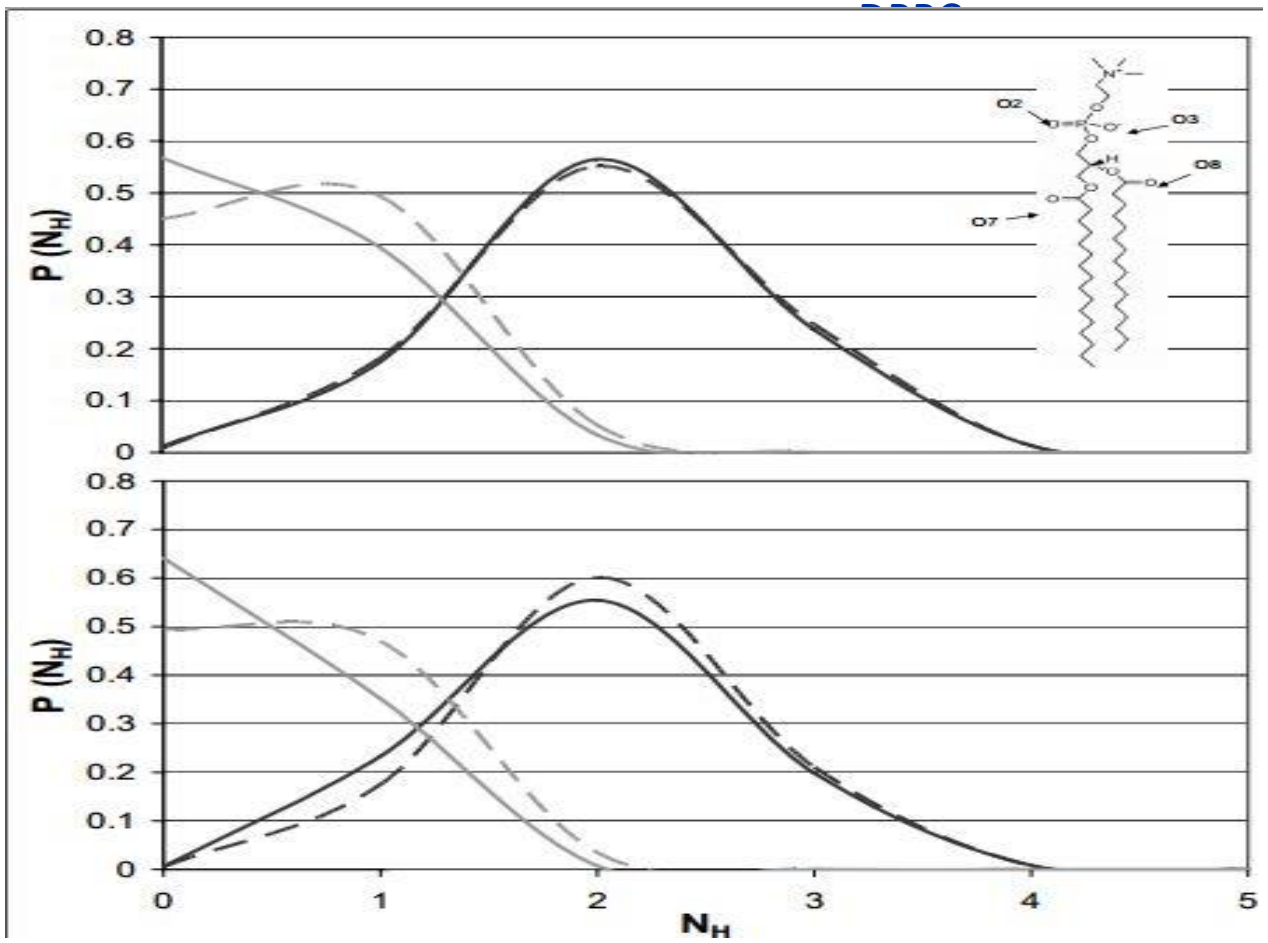




**The sequence is called WPC, where the three digits indicate the number of H-bonds it forms with other water molecules, PO and CO, respectively.**

**For instance, 100 denotes one H-bond with water and none with PO and CO, 210 indicates two H-bonds with water, one with PO and none with CO, 301 means three H-bonds with water, none with PO and one with CO, and so on. Black bars: Liquid crystalline state; light gray bars: Gel state. a) For the whole hydration water. b) For the water molecules hydrating the phosphates. c) Water around the carbonyls. d) Buried water (beyond the carbonyles).**

# Looking at hydrogen bonds we observed that,

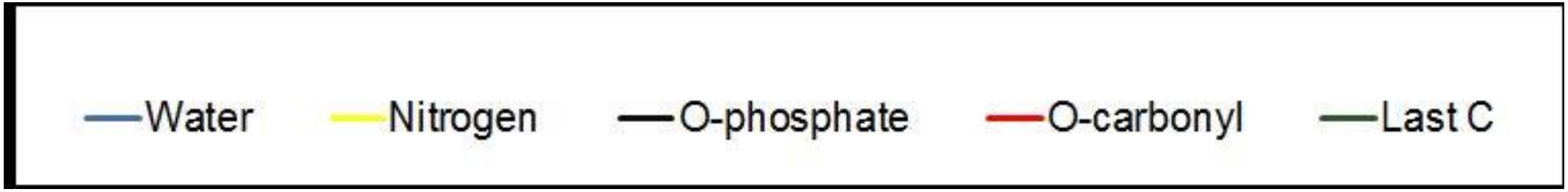


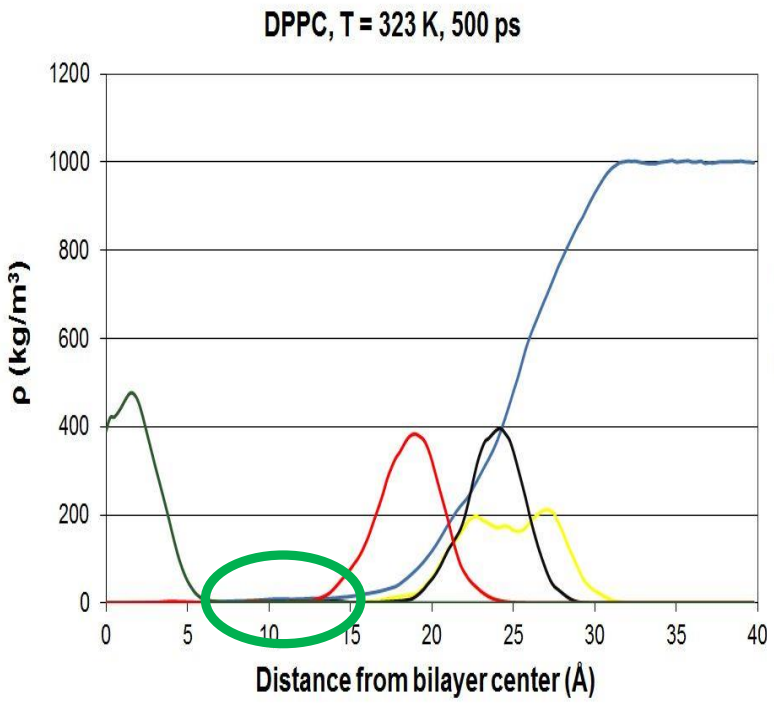

Distribution of H bonds around the different head groups (P(NH)) in the liquid crystalline (top) and gel (bottom) phases: Dark continuous line for O3, dark dotted line for O2, gray continuos line for O8 and gray dotted line for O7 groups of atoms.

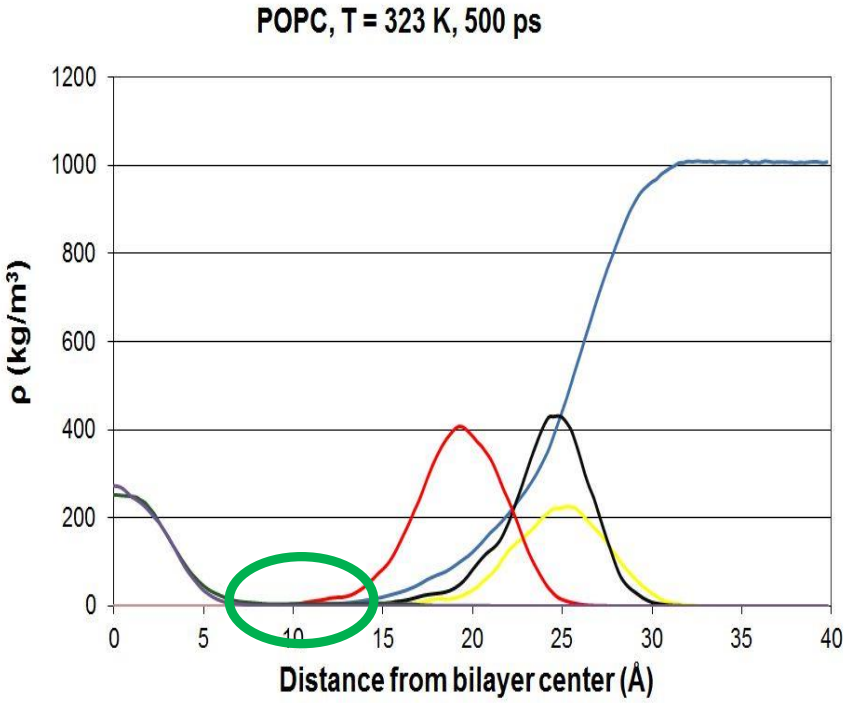
Both oxygens of the phosphate group of the lipid chain saturate their H-bond capability and possess two kinds of water molecules: directly bound by H bonds and non-bonded molecules.

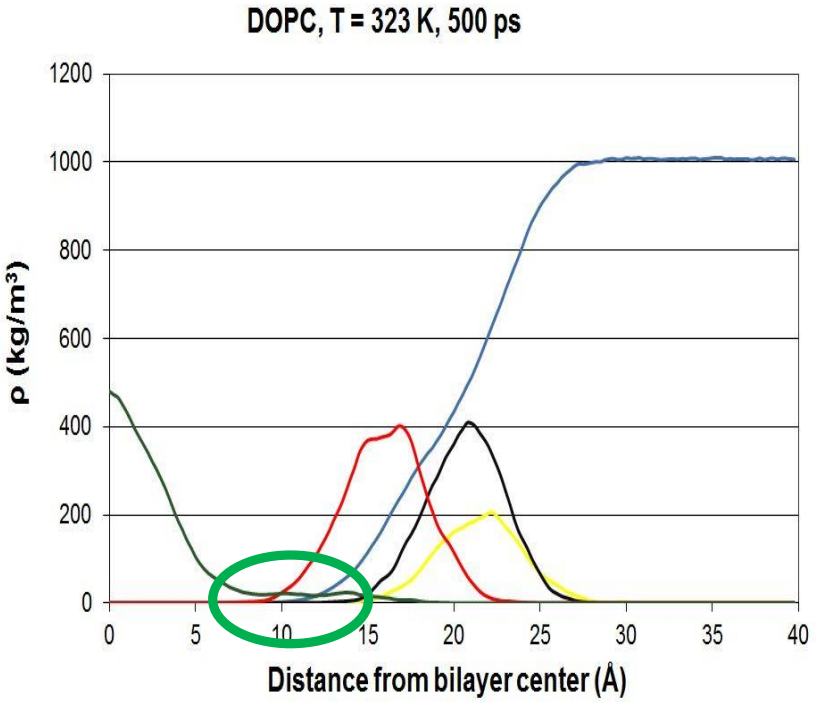
In turn, the carbonyl group of the sn2 chain forms only one H bond with water molecules, while the CO of the sn1 chain is mostly not hydrogen bonded and thus, is not significantly hydrated. This is consistent with experimental findings.

# Density Profiles



# Hydration map of DPPC and DOPC

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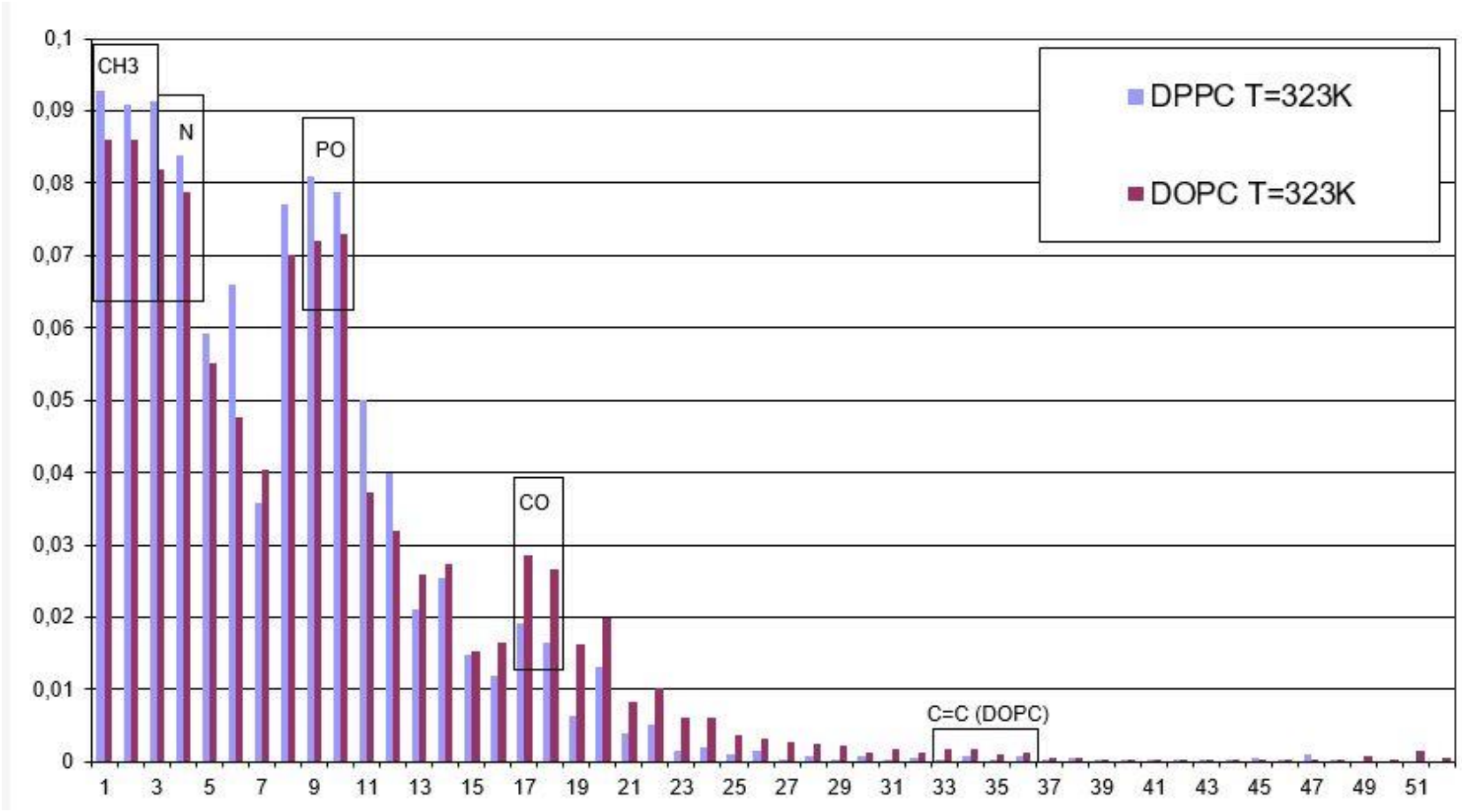


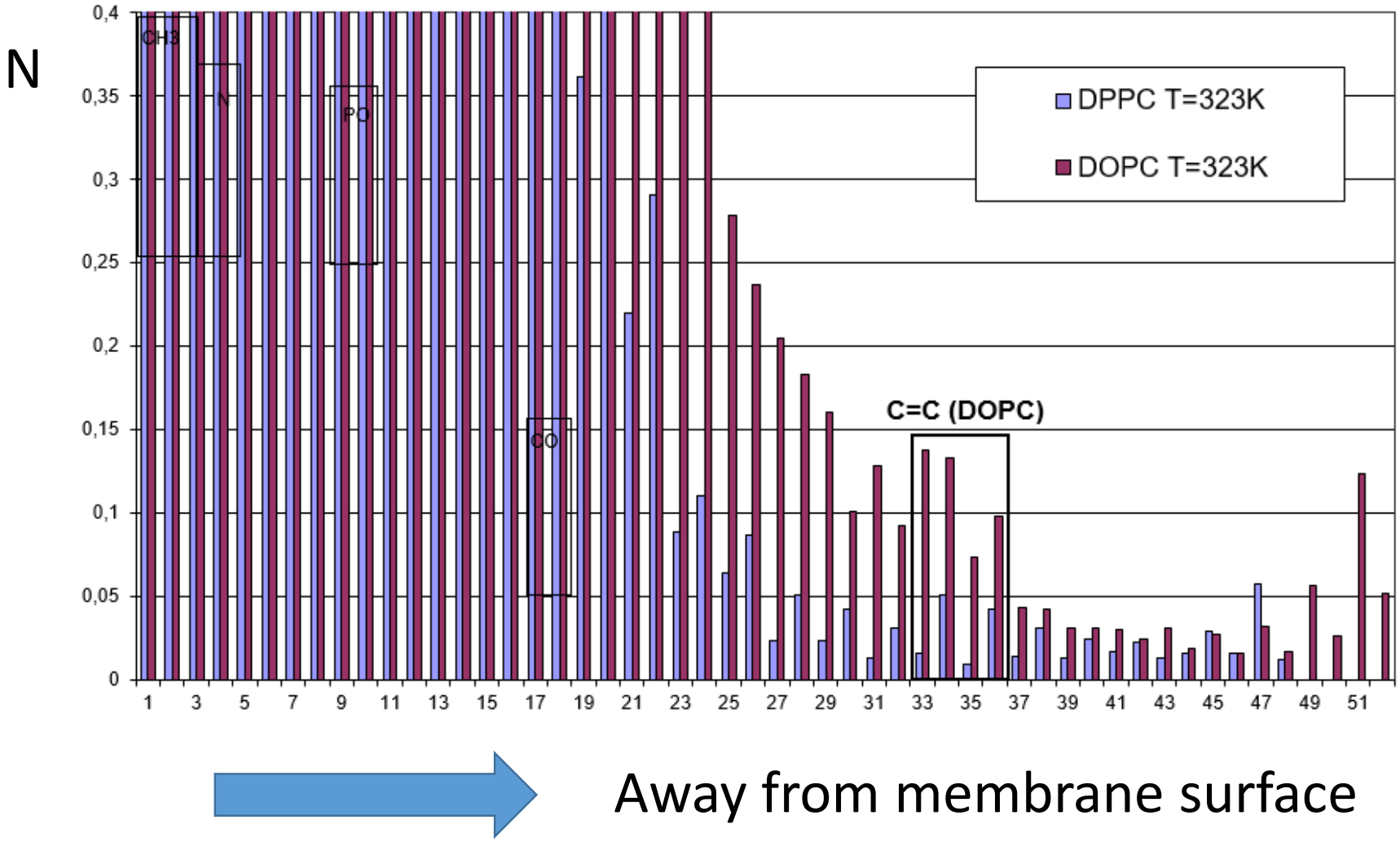

Away from membrane surface

We show the number of water molecules in a 4.5 Å radius from each lipid atom (except hydrogens).

Values shown are normalize ( the total number of water molecules per lipid is 1)

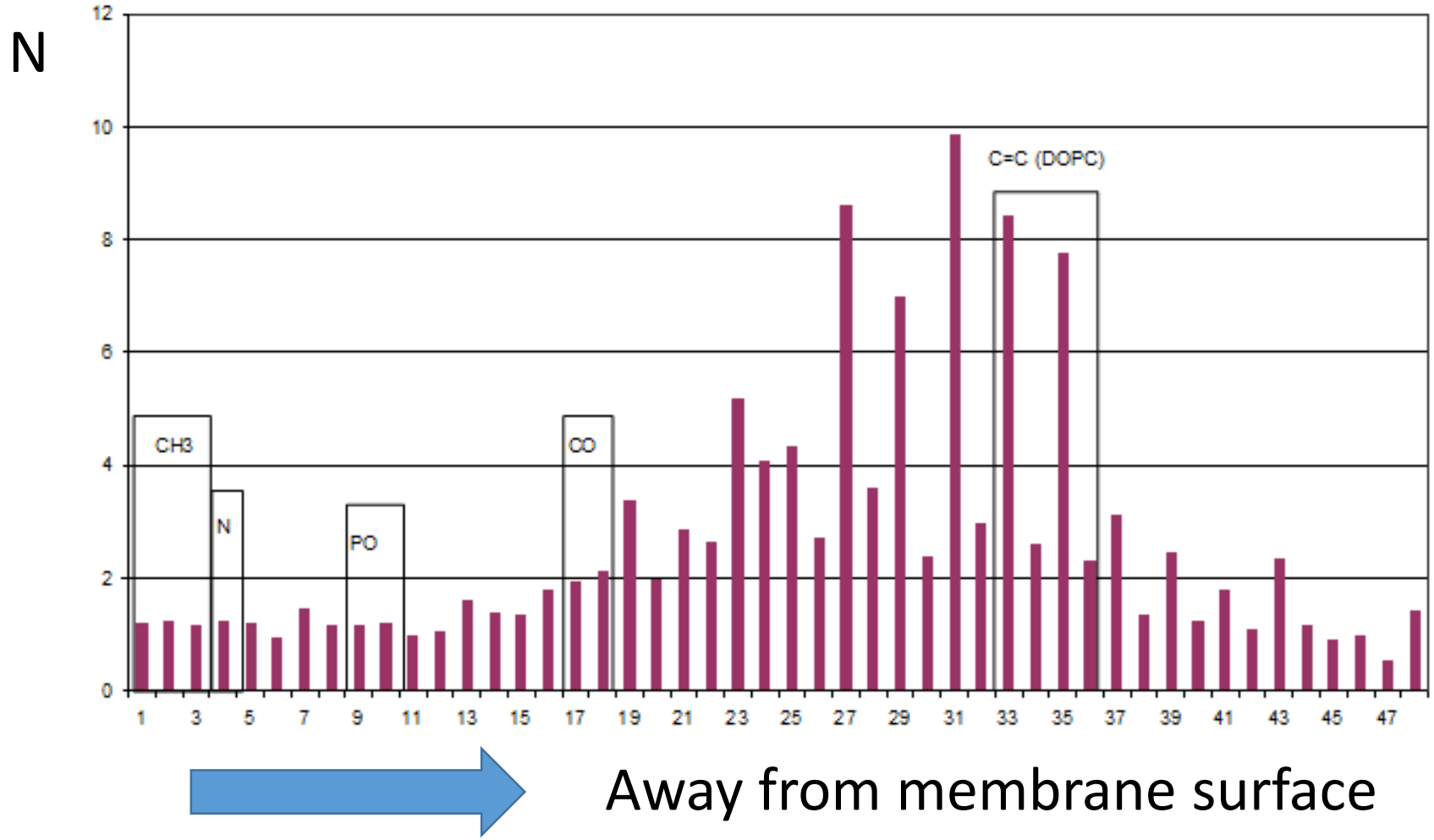
Values are higher for DPPC in the choline zone but lower in the carbonyles zone and in the interior of the membrane, compare to DOPC.

# This hydration map in a different scale (for a better description of the non-polar zone)



# Relationship DOPC/DPPC

We can see how the relationship between the water molecules of this two lipids grows as we get in the carbonyle zone and in the insaturated zone.



# Conclusions:

- We made a usefull water classification, according to number and kind of hydrogens bonds that water forms with other water molecules, PO and CO.
- In DPPC lipid bilayer we compared the membrane hydration in both phases, liquid crystalline and gel, showing the differences in the number of water molecules in several zones. Thus, the number and kind of hydration bonds in the phosphate and carbonyl zone.
- The number of water molecules in DOPC is greater in the carbonyl zone and in the insaturated zone than in DPPC. This can be interpreted from the density profiles and from the hydration maps.