

Physicochemical properties of Water and hydrophobicity in living systems

Objective:

- Introduction and Structure of water molecule
- Water and human body
- Physical and chemical properties of water
- Interactions between water molecules and hydrogen bond
- Inter-conversion between states of water
- Ice and its properties
- Polar nature of water and Universal solvent
- Amphoteric nature, Redox reaction, hydrolysis, hydrate formation
- Hydrophobicity and its biophysical effects
- Hydrophobicity and Disease states

Introduction

In this module, the fundamental properties of water, solutions will be discussed. Also the reader will be introduced to the commonly expressed terms and their units etc. Water makes up 70% of the human body and 95% water is present in plants. It is a crucial compound for the survival of all life forms. Water is the best solvent in the cell, also known as 'universal solvent', maintains the three dimensional structure of biopolymers. It is an integral part of membranes and numerous structural components. Water also maintains the physical properties of cytoplasm. And it acts as a dispersion medium near the surfaces of the macromolecules suspended in the cell and water.

Water has important effects on all biological systems. Inside each cell, there exist a number of molecules starting from simple molecules like salts and metabolites to complex macromolecules like nucleic acids, proteins etc. A plethora of molecules are 'suspended' in the biological solvent, "Water". It is well known that water makes up 80 percent of the mass of a living cell and is essential for all life processes. It is involved in every biochemical reaction, in the diffusion of nutrients into the cell and transport from the cell etc.. As seen in electron micrographic pictures of living cells, every "space" is occupied by water and

there are no empty spaces. In short, water is an integral part of the structure of the cell and some of the interesting and important facts about water in the cell are:

- Water is the best solvent in the cell, also known as ‘universal solvent’,
- It maintains the three dimensional structure of biopolymers.
- It is the integral part of membranes and numerous structural components such as microtubules and vacuoles.
- It maintains the physical properties of cytoplasm (ex. viscosity, electrical conduction, heat capacity, freezing point, and motility)
- It serves as the dispersion medium near the surfaces of the macromolecules suspended in the cell and water seems to have a longer residence time and moves more slowly.

To understand why water is so essential to life processes, it is important to understand the molecular structure of water. Pure water in itself is a fascinating chemical with a number of unusual physical and chemical properties which makes it so special. The characteristic properties of water are given below:

- Water is easily dissociated into its constituent ions, hydrogen and hydroxyl ions and back again.
- The partial charge across the water molecules makes it an excellent solvent.
- exhibits high conductivity ($\kappa = 80$), a property which allows it to act as both a reactant and a solvent.
- has a high heat capacity (C_p),
- has a high surface tension, $\sigma = 0.073 \text{ N/m}$
- has a very high boiling point (100°C)
- has a very low freezing point
- solid form of water, ice is less dense than liquid water, which is again an unusual property
- increase in fluidity with increasing pressure, and can be compressed at a constant temperature.
- one of the every few inorganic chemical which exists as a liquid at the temperature of life processes (37°)
- has a Dielectric Constant is, D of 79
- has a Dipole Moment, μ of 1.85

(i) Water structure

The key to understanding of all the unique properties of water mentioned above is the ‘hydrogen bond’. Water, (H_2O), is composed of one oxygen atom and two hydrogen atoms. Oxygen is an "electronegative" atom compared with hydrogen. Each hydrogen atom is covalently bonded to the oxygen via a shared pair

of electrons. There are four pairs of electrons surrounding the oxygen atom, two pairs involved in covalent bonds with hydrogen, and two unshared pairs on the opposite side of the oxygen atom .

Electronic Structure of Water

- Oxygen has the following electronic configuration: $1s^2 2s^2 2p^4$.
- The 2s and 2p orbitals form four sp^3 hybrid orbitals.
- These orbitals are tetrahedral in shape (bond angle distorted to 104.5°).
- The orbitals are populated such that two orbitals are filled and two contain one electron.
- The half-filled orbitals participate in the formation of a sigma bond between oxygen and hydrogen.

The two atoms of hydrogen are at an angle of 104.5 degrees from each other (i.e. **not** 180° degrees) and at a distance of 0.96 angstroms from the oxygen atom (**Fig 3.2**). The resulting molecule is asymmetric, and results in an electric dipole, causing one end of the molecule to possess a net negative charge (the oxygen end) and the other end a net positive charge. **Hydrogen bonds are strongly directional** and tend to point directly

along a line between the hydrogen nucleus and the electronegative atom.

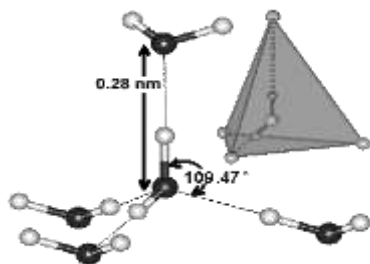


fig 3.2. Tetrahedral structure of water

Dipole is formed across the water molecule as a result of the polar covalent bonding between hydrogen and oxygen. Because the bonding electrons are shared unequally by the hydrogen and oxygen atoms partial negative charge (δ^-) forms at the oxygen end of the water molecule, and a partial positive charge (δ^+) forms at the hydrogen ends. Since the hydrogen and oxygen atoms in the molecule carry opposite (though partial) charges, nearby water molecules are attracted to each other like tiny little magnets. The electrostatic attraction between the δ^+ hydrogen and the δ^- oxygen in adjacent molecules is called **hydrogen bonding**.

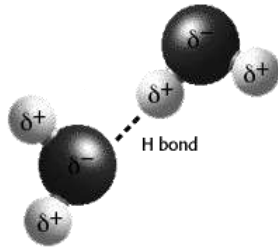


Figure 3.1: Polar nature of water and hydrogen bond formation between two water molecules

Dipole moment (μ) reflects the charge distribution of a molecule. It is defined by the following equation:

$$\mu = \sum q r \quad \dots\dots (3.1)$$

Where q is the charge on ion and r is the distance between the charges.

A larger dipole moment means that the solvent molecules can interact favorably with charged solute molecules, thus screening their charges. Consequently, a high dipole moment, D (80) usually implies a high dielectric constant. A high dielectric constant, such as that found in water, is important because the forces between charges are attenuated. As a consequence of the dipole, adjacent molecules of water may be weakly bound together, via hydrogen bonds, giving liquid water structure, particularly at low temperatures

The structure of pure liquid water consists of a random network of water molecules connected to each other by bonds from the hydrogen on one molecule to the oxygen of the next. In liquid water each molecule is hydrogen bonded to approximately 3.4 other water molecules. While, ice has a rigid lattice structure.

In ice, each water forms four hydrogen bonds in 4 other H_2O molecules with O---O distances of 2.76 \AA to the nearest oxygen neighbor. The O-O-O angles are 109 degrees, typical of a tetrahedrally coordinated lattice structure. The density of ice is $0.931 \text{ gm/cubic cm}$ as compared to the density of 1.00 gm/cc . for liquid water. The water structure in the solid ice crystal actually has vacant spaces in it . Because of which there are fewer number of water molecules in ice compared to the same volume of liquid water. In other words, ice is less dense than liquid water and will float on the surface of the liquid. Significance of hydrogen bond which explains a) solvent properties of water, b) density of ice c) ice-like structure which is present up to the boiling point, d) high boiling point e) Heat capacity and f) regulation of temperature.

Physicochemical properties of Water

1. At standard temperature 273.15 K (0 °C, 32 °F) and pressure of 100 000 Pa (1 bar water is tasteless odorless, and colorless.
2. Water has a maximum density at 3.98 °C (39.16 °F) and becomes less dense at its solid state ice. Water occupy a 9% greater volume than ice, which accounts for the floating of ice on liquid water.
3. Water is a polar molecule with an electrical dipole moment. One molecule of water can form four intermolecular hydrogen bonds that results in strong attractive forces between molecules of water, giving rise to high surface tension and capillary forces.
4. Pure water has a low electrical conductivity, which increases with the addition of ionic material.
5. The boiling point of water depend on the ambient pressure. At high altitude the boiling point decrease and near geothermal vents deep inside oceans the boiling point of water can reach temperatures of hundreds of degrees and remain liquid.
6. Liquid water can be split by electrolysis of water into hydrogen and oxygen. The energy required to split water by electrolysis is greater than the energy of formation of water when the hydrogen and oxygen recombine.
7. Water exhibit cohesion and adhesion property. Cohesion among water molecules plays a key role in the transport of water against gravity in plants. Adhesion causes capillary action.
8. Water has a greater surface tension than most other liquids because hydrogen bonds among surface water molecules resist stretching or breaking the surface. Water behaves as if covered by an invisible film. Some animals can stand, walk, or run on water without breaking the surface
9. Water has high specific heat which is the amount of heat required to raise or lower the temperature of 1 gram of substance 1 degree C. In other words, water can absorb a lot of heat with little change in temperature. Because of this water acts as a thermal buffer and has high thermal capacity. Water stabilizes air temperatures by absorbing heat from warmer air and releasing heat to cooler air.
10. Water has high Heat of vaporization which is the amount of heat required for 1 gram of a substance to be converted from a liquid to a gas. It helps our bodies and our planet to maintain our temperature within a tolerable range. When we get hot and sweat, water evaporates from our skin and cools us. Since the evaporation of water requires a considerable amount of energy, it is very effective in cooling us.

11. Water is highly dense at 4°C and contracts until 4°C and expands from 4°C to 0°C. The density of water: 1) Prevents water from freezing from the bottom up. 2) Ice forms on the surface first—the freezing of the water releases heat to the water below creating insulation. 3) Makes transition between season less abrupt.
12. The highly polar nature of water leads to hydrogen bonding which is the maximum in ice and least in water vapour. The polar nature of water also makes it: (a) a very good Solvent (b) to act as an amphoteric substance and (c) to form hydrates of different types.
13. Water is capable of acting as either an acid or a base and can undergo self-ionization. The self-ionization of water can be expressed as:
- $$\text{H}_2\text{O} + \text{H}_2\text{O} \rightleftharpoons \text{H}_3\text{O}^+ \text{aq} + \text{OH}^- \text{aq}$$
- acid-1 base-2 acid-2 base-1
- (acid) (base) (conjugate acid) (conjugate base)
14. Decomposition of water is a redox reaction. The oxidation reaction occurs at one electrode, and the reduction reaction at the other electrode.
- Water is oxidized at the anode. The reaction is...
- $$2\text{H}_2\text{O} \rightarrow \text{O}_2 + 4\text{H}^+ + 4\text{e}^-$$
- Water is reduced at the cathode. The reaction is...
- $$4\text{H}_2\text{O} + 4\text{e}^- \rightarrow 2\text{H}_2 + 4\text{OH}^-$$
15. Water has a high dielectric constant and has a strong hydrating tendency. It dissolves many ionic compounds. However, certain covalent and some ionic compounds are hydrolysed in water. Also water can crystallize many salts from aqueous solutions as hydrated salts.
16. Water is a good polar solvent and a *universal solvent*. Substances that dissolve in water, are known as *hydrophilic* substances, while those that are immiscible with water are called as *hydrophobic* substances. All cellular components are dissolved in water to derive their structure and activity. Moreover water forms a homogeneous mixture with many liquids, such as ethanol, in all proportions but usually form a layer with the least dense liquid as the top layer, and the most dense layer at the bottom when mixed with oils.

(ii) Bio-water

Water can hydrogen-bond not only to itself, but also to any other molecules that have -OH or -NH₂ units such as alcohols, and macromolecules such as proteins etc.. The biological activity of proteins is critically dependent not only on their composition but also on the way they are folded; this folding involves

hydrogen-bonded interactions. The asymmetric distribution of solutes can be explained by preferential binding and the partition coefficient of the solute between the intracellular-water and extra-cellular water. Cell in a highly integrated system of multiple components interacting with one another and maintains its physical integrity, ion exclusion properties. Cell water in the vicinity of surfaces and macromolecules is in ion- extended hydration shells. Near the surfaces of the macromolecules suspended -water may have a longer residence time So it is, in effect, a different kind of water, a different kind of solvent, near the surfaces. But away from the surfaces and charged groups, the cell water begins to act more like free water. This argument can explain differences in the physical properties of water in the cell while the membrane theory would not.

It is now known that the intracellular water, which is very close to any membrane or organelle (sometimes called *vicinal water*) is organized very differently from the bulk water, and that this structured water plays a significant role in governing the shape and thus biological activity of large folded biopolymers. It is important to bear in mind, however, that the structure of the water in these regions is imposed solely by the geometry of the surrounding hydrogen bonding sites. Data from several techniques, NMR, ESR, calorimetry and quasielastic neutron scattering has proved that the ‘residence time’ of the water molecules is increased and the ‘relative motional freedom’ of water is decreased inside cells. Cancer cells have peculiarities of salt and water balance and cancers are simply have more water or “wetter “than the normal cells. Extended fibrous proteins may have preferential binding sites for potassium and may also affect the ability of water molecules in their vicinity to dissolve ions and other small molecules (Presence of non-polar molecules, makes water molecules pack into ordered configurations and formation of the ice-lattice formation).

(iii) Structure-making and Structure-breaking

The behavior of aqueous solutions of small and large molecules is influenced considerably by the nature of the solvent itself. Water serves as the major hydrogen donor in enzyme reactions, and ions travel through the cell in a hydrated form, the diameter of which is important in ion discrimination. The ability of ions and other molecules to dissolve in water is due to polarity. Pure water ionizes to a very small degree through to a measurable extent. Structure-making ions like, calcium Ca^{+2} , lithium Li^{+} , sodium Na^{+} , form stronger bonds with water, decrease free movement of water molecules and thereby increase hexamer structure formation. ‘Structure-breaking’ ions like magnesium Mg^{+2} , potassium K^{+} , form weaker bonds with water, therefore increase free movement of water molecules and decrease hexamer structure (or increase unstructured water clusters). Therefore, calcium and sodium can be used to create hexagonal

water clusters, ions such as magnesium and potassium tend to destroy such hexagonal clusters, producing pentagonal or other unorganized water clusters.

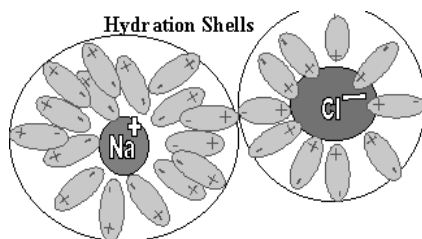


Figure 3 Presence of a salt disturbs the water structure. The water molecules reorient around the ions. This makes the ions to move with their primary hydration shells.

Hydrophobicity

Hydrophobicity is a term poorly understood rather often misunderstood term. It is not correct to say that hydrophobic residues attract each other via hydrophobic forces. There are weak attractive forces, between atoms Van der Waals interactions. The force associated with this Van der Waals interaction is very weak at large inter-atomic distances and still weak when the atoms get closer. A water molecule in liquid form makes about 3.5 hydrogen bonds with other and it has nearly 6 degrees of freedom. It is important to mention here, that freedom like rotational freedom and translational freedom contribute to the entropy and the hydrogen bonds contribute to the enthalpy in the overall free energy. Unlike polar interactions, van der Waals forces are not directional. Now an aromatic or any other hydrocarbon molecule is introduced in water, water can only form two hydrogen bonds, and its freedom is also highly restricted. Such a water molecule is therefore different from the one in bulk water.

Hydrophobicity is measured by measuring ΔG_{trans} , the transfer of say for example, amino acid from water to an organic solvent

$$\Delta G_{trans} = RT \ln(P) \text{ , where, } P = X_{aq} / X_{nonaq} \text{ and } X \text{ is mole fraction}$$

Therefore, $\Delta G_{trans} = +ve$, then the amino acid is hydrophobic, and if $\Delta G_{trans} = -ve$ then the group is hydrophilic.

Biological phenomenon where the concept of hydrophobicity plays a role

1. Protein folding: In proteins, many hydrophobic residues pack against each other in the core of the protein. Infact 'Protein hydrophobicity plot' for protein sequences are characterized for their hydrophobic character like predicting membrane-spanning domains, potential antigenic sites and regions that are likely exposed on the protein's surface. Since water is an important factor in determining the

energy balance during protein folding, **hydrophobic effect** plays a crucial role in dictating the native conformation of a protein.

2. Compartmentalization in the cell: Cell organelles are surrounded by flat sheets of bilayer phospholipid membrane that form a barrier and is called as COMPARTMENTALIZATION. The hydrophilic heads face and interact with water whereas the hydrophobic tails form the interior of the bilayer. This helps different organelles to maintain concentrations of substances, maintain pH and regulate specialized functions efficiently

3. Drug delivery: Liposome is a spherical molecule surrounded by hydrophobic lipid bilayer, and can contain molecules at center, enabling them to deliver drug. Liposome carries such drugs into body and fuses with the other membranes to release the drug at the site.

4. Hydrophobicity and disease: Sickle cell anemia: Autosomal recessive disease that causes abnormality in the oxygen-carrying protein haemoglobin. Caused by point mutation in the β -globin chain. Position 6 is located at the surface of the beta chain, where it would normally be exposed to water. At this position hydrophilic GLUTAMIC ACID gets replaced with hydrophobic VALINE. Switch from a hydrophilic to a hydrophobic residue on the surface reduces solubility of the molecule and promotes the formation of large insoluble aggregates

5. Alzheimer's Disease: Gamma and beta secretases cleave Amyloid Precursor Protein to release a 42 amino acid peptide called as amyloid beta fragment. This then aggregates in the hippocampal region, mainly owing to the hydrophobic nature. This is the pathogenesis of Alzheimer's disease.

Summary:

- ❑ Water is the most abundantly available substance of a great biological and chemical significance.
- ❑ Ease with which water is transformed from liquid to solid and to gaseous state allows it to play a vital role in the biosphere.
- ❑ The physical property of water include (1) Cohesion (2) Adhesion (3) High Specific Heat (4) High Heat of Vaporization (5) low density of ice.
- ❑ Highly polar nature of water leads to hydrogen bonding which is the maximum in ice and least in water vapour.
- ❑ Polar nature of water also makes it: (a) a very good; Solvent (b) act as an amphoteric substance and (c) forms hydrates of different types.
- ❑ Hydrophobicity is the physical property of a molecule that is repelled from water
- ❑ Hydrophobicity has a role to play in protein folding & stability, Compartmentation, drug delivery and causation of sickle cell anaemia and Alzheimers disease