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NANOSCALE MASS TRANSPORT IN LIQUIDS

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ABSTRACT

Intermolecular forces in liquids — van der Waals, electrostatic, steric — range over a distance of about 1-100 nm. When liquids are confined to such length scales, their properties and behavior can be significantly altered. In this paper, I have discussed how confined liquids can be used to study biomolecules and biomolecular reactions. Much of the fundamental knowledge is yet to be developed, although this may have direct impact on biotechnology.

INTRODUCTION

It is fair to say that over the past decade, micro/nanoscale heat transfer has been perhaps the most exciting and active area of research within the heat transfer community, and has also attracted the most amount of research funding. Activity in this area started sometime in the late 1980s. The research was partly driven by the tremendous success of microelectronics and a clear vision that devices were going to become smaller and faster, and that future developments would require an extensive knowledge base for the continuously shrinking time and length scales that would reach tens of nanometers within decades. This also coincided with the invention of a wide variety of experimental tools such as the scanning tunneling and force microscopes and femtosecond lasers, which provided immediate access to phenomena at nanoscales. The ability to make microdevices using MEMS technology became standardized at around the same time. In addition, computing power became readily available such that molecular dynamic and stochastic simulations of micro/nanoscale thermal phenomena became feasible. All these factors put together led to an intense influx and genesis of new theoretical concepts, experimental techniques, and device designs, which led to a remarkable pace of progress. While the research activity continues to grow, one may ask if there is anything left to do or if there are research areas that we should be exploring in the future.

I argue in this paper that while micro/nanoscale heat transfer has received much attention, the fundamentals of nanoscale mass transport, especially in reacting flows, have not been fully explored. Yet they are found in energy conversion devices such as fuel cells. Furthermore, molecular biology provides many examples were mass transport and reactions at nanoscales are not only important for life processes, but a

Table 1 Characteristic length and time scales for energy carriers under ambient conditions.

Energy Carrier ⇒ Characteristics⇓	Molecules in Gas	Molecules in Liquid	Electrons in Metal and Semiconductors	Phonons in Dielectric or Semiconductor
Mean Free Path, ℓ [nm]	100	0.1-1	1-50	1-500
Wavelength, λ [nm]	< 0.1	< 0.1	0.1-50	0.1-5
Relaxation Time, τ [s]	$(0.1\text{-}1)\times10^{-9}$	$(0.1\text{-}1)\times10^{-12}$	$(10\text{-}100)\times10^{-15}$	$(1\text{-}10)\times10^{-12}$
Propagation Speed, v [m/s]	$(0.2\text{-}2)\times10^{3}$	$(1\text{-}5)\times10^{3}$	$\approx10^{6}$	$(3\text{-}10)\times10^{3}$
Equilibrium Statistics	Maxwell-Boltzmann	Maxwell-Boltzmann	Fermi-Dirac	Bose-Einstein

fundamental understanding can lead to new technologies. So what is unique about micro/nanoscale mass transport in reacting flows?

Table 1 provides some characteristic length and time scales for transport phenomena in solids, liquids and gases. For solids and gases, the mean free path is one of the fundamental length scale that demarcates the transition between classical and microscale transport phenomena. For electrons and phonons in solids, the wavelength of energy carriers is also important and lead to the quantum mechanical aspects of transport. Table 1 provides the mean free path of molecules in liquids, which is on the order of 0.1-1 nm. Given this, one would expect no transitions in behavior until one reaches these length scales. It is important to note, however, that the mean free path is not the characteristic length scale that determines transitions in a liquid's behavior. The thermophysical and transport properties of liquids are determined instead by intermolecular and surface forces. So what are these forces and what are the range of length scales that they operate in?

NOMENCLATURE

f_o = equilibrium statistical distrubution
k_B = Boltzmann constant
ℓ = mean free path
ℓ_D = Debye length
T = temperature
ε = dielectric constant
ε_o = permittivity of vacuum
η = ion concentration
ρ = electrical resistivity
τ = relaxation time

INTERMOLECULAR FORCES IN LIQUIDS

When a liquid is in close proximity to a solid surface or a biological molecule, there are four types of forces that are significant - hydration, electrostatic, van der Waals forces, and elastic strain.

Hydration Forces

Solid surfaces such as silica, mica or any other oxides or biological molecules such as DNA or proteins that contain a polar group, are hydrophilic and form hydrogen bonds with water. The H-bond strength between water molecules and the surface can be so strong that the first few layers of water completely wet the surface and are often highly structured. Experimental evidence of such highly ordered 2-dimensional phase of water has been obtained by several investigators [1], which suggest that the strong binding energy decays exponentially away from the surface with a characteristic length scale of 1-2 nm.

Electrostatic Forces

A solid surface or a biological molecule can be charged in two ways: (i) by ionization or dissociation of a surface group - for example, removal of H^+ ions from a glass surface; (ii) adsorption of ions from solution to a previously uncharged surface - for example adsorption of Ca^{2+} ions from solution to replace the K^+ ions in mica. When a surface or molecule is charged, a region of oppositely charged counterions in solution are attracted to the surface. The charge density of counterions decreases exponentially with distance from the surface and approaches the bulk value far away. The combination of surface charge and counterions in solution forms a region near the surface called the *electric double layer*. The length scale that characterizes the exponential decay of electrostatic forces is called the Debye length, ℓD, which can be expressed as

$$\frac{1}{\ell_D} = \sqrt{\sum_i \frac{\eta_{\infty i} e^2 v_i^2}{\varepsilon \varepsilon_o k_B T}} \qquad (4)$$

where ε is the dielectric constant of the medium, ε_O is the permittivity of vacuum, e is the charge of an electron, vi is the valence of the ith specie ion and $\eta_{\infty i}$ is the concentration of the ith ion specie far away from a surface. Typically, ℓD is on the order of 1-50 nm depending on the ion concentration in the bulk.

van der Waals Forces

Although not as strong as hydration or electrostatic forces, van der Waals forces are always present whenever a liquid is adsorbed onto a surface. They arise from interactions of induced dipoles between two or more atoms and are typically effective below 1-50 nm.

Elastic Strain

The fact that water molecules are highly ordered in a two-dimensional ice-like structure close to a hydrophilic surface has been established through experiments and molecular dynamic simulations. Therefore, if the lattice constant of the substrate is not exactly equal to that of ice, the bonds between the water molecules will be strained. The fact that strain influences phenomena at the solid-liquid interface has been known for a while. For example, silver iodide (AgI) has been used to nucleate ice and it is commonly believed that it is the lattice match of the crystal structure of AgI and ice that promotes ice nucleation [2]. Experiments have also shown that strain energy can play a significant role in nucleation processes, in particular for heterogeneous nucleation of ice from water on a solid surface [3,4,5].

LIQUID TRANSPORT IN CONFINED REGIONS

It is clear from the above discussion that all the surface forces in liquids operate at different length scales — some are near field and others far field. It is important to note, however, that regardless of their origin, they all operate in the range of 0.1-100 nm. Hence, it is only when a liquid is confined to 100 nm and below by solid walls that one should expect to observe changes in transport phenomena. It must be recognized that the increased surface-to-volume ratio of nanostructured solids is widely used to make compact catalytic reactors. An application of such reactors are in fuel cells, where nanoparticle catalysts improve efficiency. In this paper, I will not discuss transport in catalytic reactors, but focus on nanoscale transport phenomena in molecular biology.

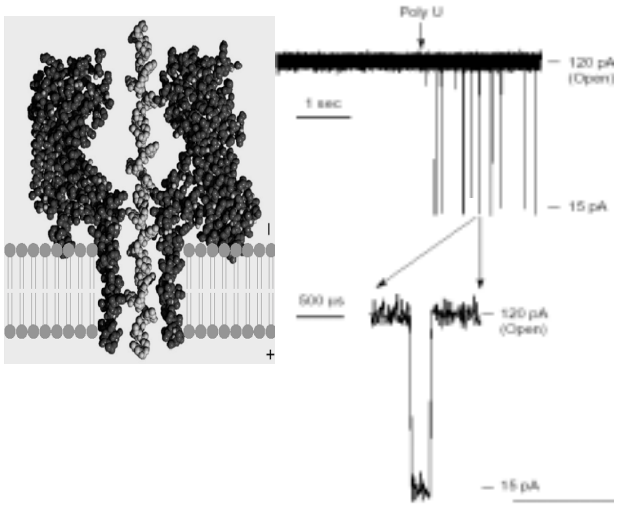

Figure 1 A 120 mV bias across αHL ion channel produces an ionic current of ≈ 120 pA. When a single polynucleotide strand passes through the channel, the current drops to 15-50 pA. The amplitude of the current drop and its duration depend on the type of nucleotide [9].

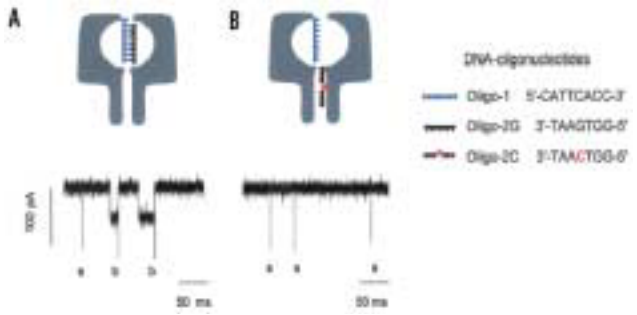

Figure 2 A probe ssDNA is attached through a disulphide linkage to a cysteine residue at the cis opening of a αHL nanopore protein, as schematically shown above [15]. Shown below are the time traces of the ionic current passing through the nanopore. If the complementary target ssDNA is transported through the nanopore, it binds with the probe strand, which reduces the ionic current for approx. 50 ms. However, if a single base pair mismatch is introduced the binding lifetime is reduced to about 1 ms.

Biological Nanopores

Recent work has shown that by monitoring the ion current across a nanopore (1.5-4 nm inner diameter) formed by transmembrane protein ion channels, it is possible to detect single-molecule binding events with the specificity of single base pair mismatches of DNA. The work can be broadly classified into two categories, namely: (i) non-functionalized nanopores; (ii) functionalized nanopores. Almost all of the past work has involved the transmembrane protein ion channel α-Hemolysin (αHL) (see Fig. 1) embedded in a suspended membrane separating two chambers filled with ionic solution. The entrance on the top (*cis*) side is about 2.6 nm in diameter whereas the narrow channel through the membrane that is closer to the bottom end (*trans*) is 1.4 nm in diameter. When a voltage bias [6] of 120 mV is applied across the ion channel, an ionic current of about 120 pA is produced for ionic concentrations of 1 M KCl (the resistance is approximately 109 Ω). When single-stranded polynucleotides are introduced in one of the chambers, they electrophoretically flow through the ion channel. By doing so, they block the ionic current, which reduces to levels of 15-50 pA. This is because the size of the polynucleotides is on the order of the pore size. The time of flight of these polynucleotides seems to vary linearly with their length, and inversely with the applied voltage. It has been hypothesized that different nucleotides would have different blocking signatures (either time of flight or amplitude of current drop), which would allow one to rapidly sequence single-stranded DNA (ssDNA) directly [7]. This has led to many attempts over the last decade, and there has been partial success in discriminating between different bases [8]. For example, polyCs seem to produce shorter but deeper (lower current) decrease in ionic current whereas polyAs produce longer but shallower reductions [9]. However, direct and rapid sequencing of ssDNA has been unsuccessful and remains a challenge, although hairpin DNA molecules have been detected with single nucleotide resolution [10]. The problem in direct sequencing arises from the fact that the time a single base spends in the nanopore is too short and that the number of ions that it blocks is too few (approx. 100), making it difficult to detect it above the noise. Slowing down the polynucleotides could offer a chance of direct sequencing, but that has also remained a challenge.

While it has so far been very difficult to achieve biomolecule specificity using non-functionalized nanopores, recent work using functionalized αHL nanopores has been most promising. Howorka et al. [11] functionalized the nanopore with a ssDNA probe attached at the *cis* entrance through a disulphide linkage to a cysteine residue in the αHL protein (see Fig. 2). Then by transporting target ssDNA sequences, they found that when the target was fully complementary, its residence time in the nanopore, as measured by the duration of the reduced ionic current, was much longer (≈ 50 ms) than if even a single base-pair mismatch is introduced (≈ 1 ms). From this, kinetics of the binding reaction can be quantified [12]. Furthermore, the Bayley group [13,14,15] has also functionlized αHL protein nanopores with other molecules to study reaction kinetics of various molecular interactions such as small molecules with proteins, ions with proteins, etc.

Synthetic Nanopores and Nanotubes

While biology can provide exquisite control on the pore structure and chemistry, they are difficult to integrate them on Si or other inorganic platforms. Hence, synthetic ones made of inorganic materials are desirable. However, fabricating nanopores with 1-2 nm precision is extremely difficult. Recently, Li et al. [16] have demonstrated that artificial nanopores made of inorganic materials (SiNx) can be fabricated using ion beam sculpting, and that they show similar behavior in blocking ionic current when ssDNA passes through them(see Fig. 3). However, direct sequencing of ssDNA has not been

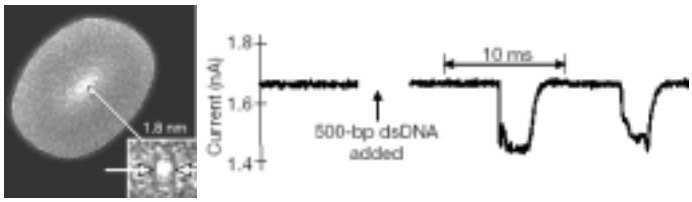

Figure 3 Synthetic nanopore fabricated in SiN_x by ion sculpting [16]. Passage of a 500bp double-stranded DNA produces a drop in ionic current in response to a 120 mV bias across a nanopore in the presence of a 1M KCl solution.

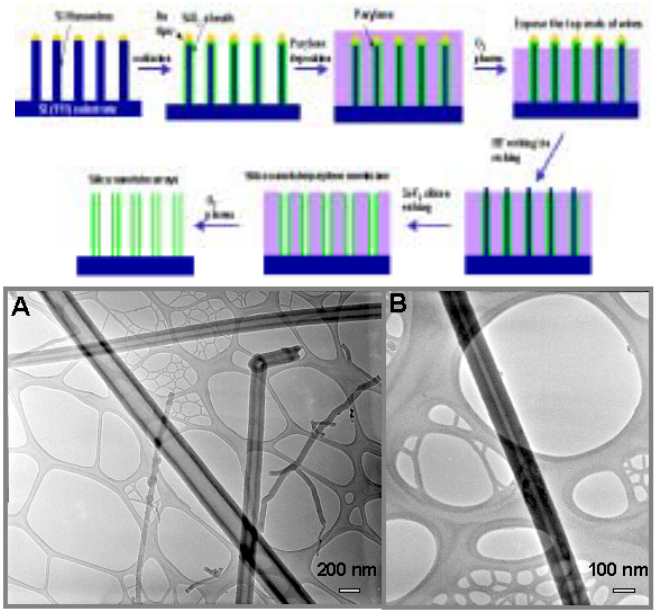

Figure 4 (Top) Flow chart of the fabrication process for making silica nanotube arrays from vertical silicon nanowire templates. (Bottom) Transmission electron micrographs for the as-prepared silica nanotubes.

reported so far and parallel processing of those artificial nanopores proves to be very difficult with Li's approach.

We have recently developed a process of converting nanowires into nanotubes [17]. Figure 4 shows the fabrication process and some SiO2 nanotubes. It must be noted carbon nanotubes, which have been widely used for nanoscale science and engineering, are hydrophobic in nature and it is very difficult to introduce water inside them. Silica nanotube are, in contrast, hydrophilic such that capillary pressure is sufficient to fill it with liquid. Hence, such nanotubes could be used to study biomolecular transport and reactions. We plan to use these nanotubes to study transport and reactions of biomolecules, perhaps single molecule at a time.

SURFACE BIOMOLECULAR REACTIONS

One of the technologies that has revolutionized the way genomics research is performed nowadays is DNA microarrays. Figure 5 shows an image of thousands of spots where fluorescently labeled target single-stranded DNA (ssDNA) have hybridized with their complementary strand that is

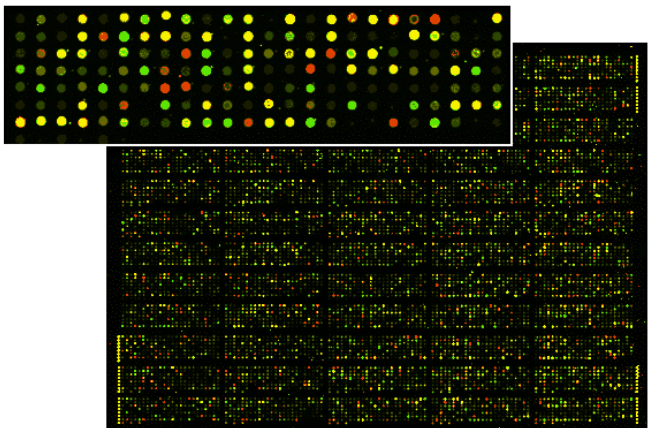

Fig. 5 Fluorescent image of a DNA microarray showing more thousands of fluorescent spots indicating DNA hybridization.

Fig. 6 Surface reaction of DNA hybridization occurs in DNA microarrays.

attached to a solid substrate (generally glass). Such gene chips allow one to simultaneously study the occurrence or mutations in thousands of genes. In addition, the expression of these genes in the form of messenger RNA (mRNA) can also be quantitatively measured, thus giving insight about the molecular machinery inside a cell. Such DNA microarrays are used not only in basic science but also in applications as well, including forensics, drug discovery, diagnostics etc.

The fundamental process that occurs in such DNA microarrays is a surface chemical reaction, as illustrated in Fig. 6. The speed and efficiency of this reaction depends on a number of factors, namely: concentration of target ssDNA, density of probe ssDNA, length of probe and target ssDNA, and ion concentration. Although DNA microarrays are widely used, to a large extent their performance, although critical, is not well understood. Georgiadis and coworkers [18,19] have performed careful experiments to study the dependence on various factors. Figure 7 shows the effect of surface probe molecular density on the hybridization efficiency. The surface molecular density varies from 2-12 x 10^{12} cm^{-2}, which relates intermolecular distance of 3-7 nm. This falls in the regime of hydration, electrostatic and van der Waals forces, such that biomolecular reactions would be significantly altered in such confined regions over that in bulk fluid. The difference in the time taken as well as the efficiency of the reaction suggests that it would be very important to control the mass transport as well as reaction kinetics. Fundamental knowledge of nanoscale fluidic interactions through changes in intermolecular forces is the key to designing such biochips with optimized performance.

While DNA microarrays are commercially produced and widely used, protein microarrays that utilize antibodies as probe molecules are starting to be developed. Here again the speed

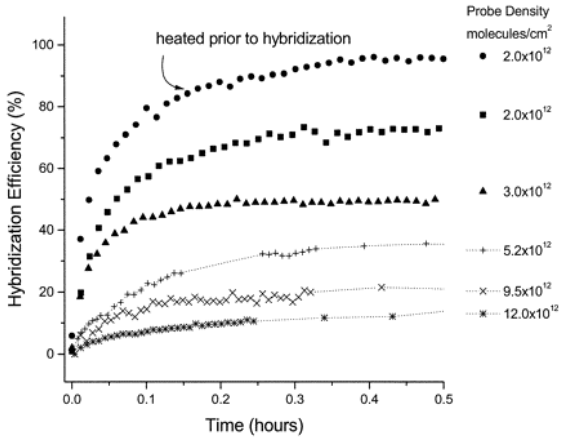

Fig. 7 Target hybridization kinetics as a function of probe density. The probe density, determined by surface plasmon resonance, varies from 2×10^{12} to 12×10^{12} molecules/cm^2. Heating of the probe film prior to hybridization increases the hybridization efficiency. All runs are 1 μM target in 1 M NaCl with buffer.

and efficiency of surface reactions would depend on the density of probe molecules, intermolecular forces between them and mass transport of target protein molecules to the surface. This would be an important and exciting area of research.

CONCLUSIONS

Most of the intermolecular forces in liquids that determine the behavior such liquids generally occur with a length scale regime of 1-100 nm. Hence when synthetic or naturally occurring structure fall in this regime, fluid behavior in such confined regions can be drastically different. In this paper, I have suggested how biomolecular transport at nanoscales can be studied and utilized to make contributions both in basic science as well as in biotechnology. I believe this is an exciting area of research where the heat transfer community, and in general researchers in thermofluid science and engineering, can make contributions.

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