Part I

Molecular forces
1 Reasons for the enquiry

1.1 Molecular forces: some of the background and history of ideas.

Why molecular forces?

The matter that concerns us was most clearly articulated nearly a century ago by D’Arcy Thompson in his famous book [1]. He reported the pleas of the early founders of the cell theory, of the then biology, and of the physiologists, that chemists should address the question of molecular forces, then unknown.

We would like to know how it is that molecular forces and the laws of statistical mechanics conspire, with the geometry of molecules and the conformations available to macromolecules, to give rise to the hierarchies of self-assembled equilibrium or dynamic steady states of matter that form cells and dictate biochemical reactivity.

In other words, the game is to link structure and function, the geometry of assemblies of molecules, to the forces that drive self assembly and recognition processes. Any insights ought to allow us to build better, useful connections between the physical and biological sciences. Despite tantalizing hints, that main aim has remained elusive.

D’Arcy Thompson tells us too that of the chemistry of his day and age, Kant said that ‘it was wisschenschaft, nicht Wissenschaft; in that the criterion of a true science lay in its reliance on mathematics’. Kant believed that Euclid’s geometry was self-evidently that of nature. We now know better. Hyperbolic geometries that we shall come to later better describe the bicontinuous, random honeycombs of nature. In apposition to Kant’s position we have it on the authority of Auguste Comte, the founder of the social sciences, that: ‘If mathematics should ever hold a prominent place in chemistry, an aberration happily almost impossible, it would occasion a widespread and rapid degeneration of that science’. The quotation is out of context and probably not fair to Comte. But these two opposing views do reflect the ambiguous position of chemistry in science. One view has it that chemistry has...
Reasons for the enquiry

to rely on physics and mathematics. The other is that this is a too simplistic, even arrogant expectation.

The word chemistry derives from Al-Khemie, itself supposed to come from a Greek word meaning the black land (renewal of silt from the Nile floods) or Egypt, from which all wisdom came. Some of the magic, mystery and art associated with medieval alchemists still attends some branches of chemistry, the science of molecules, in accordance with Comte’s view. But physics, natural philosophy, explores the properties of matter and energy, or the action of different forms of energy on matter generally, and was missing. In the endeavour, and par excellence for physical chemistry, forces and therefore mathematics have to figure centrally.

Newton himself had tried to measure the molecular forces acting between surfaces and reported in Art. 31 of the *Principia* that he failed, because ‘surface combinations were owing’. The problems of surface and colloid chemistry were already writ large.

Thomas Young deduced the range of the force between atoms and molecules, this before atoms were agreed to exist. That these forces had to be short-range, unlike gravitational forces, was also known to Newton.

The Reverend Challis of Trinity College, Cambridge, reviewed, in the 1836 meeting of the British Association for Advancement of Science, the then state of play of a dispute between Laplace and Poisson on what we might now call hydration forces at and between surfaces. George Peacock, a professor of mathematics at Cambridge and Young’s biographer, furiously reports that Laplace stole Thomas Young’s earlier work, on contact angles and surface tension, without attribution. To make it worse, while Young, a writer of great clarity, went to considerable trouble to avoid mathematics, Laplace added an appendix to his Celeste Mechanique on the topic, dressing it up in fancy mathematics. (And incidentally, Laplace got it wrong as he forgot about contact angles formed by a drop of liquid on a solid interface.) Poisson lost out because of a mistake by a factor of two, and to too scrupulous adherence to Ockam’s razor by his opponents.

This matter was redressed later in a marvellous article long forgotten on the theory of capillary action by J. Clerk Maxwell in the 1876 edition of the *Encyclopaedia Britannica*, updated by Lord Rayleigh in the eleventh edition. Poisson was right. This apparently absurd dispute between Poisson and Laplace is a conceptual matter, of confusing and central importance. The nature of an interface is with us still, indeed endemic in chemistry. In its simplest form it boils down to this: imagine an ideal, i.e., molecularly smooth surface, like mica. Then, can a liquid that adjoins it be considered to have its bulk density (or with complicated molecules such as water, random molecular orientation) up to an infinitesimal, molecular, distance from the surface (the Laplace approximation)? Or, under the influence of forces between the various constituent molecules, do the liquid molecules exhibit a
1.1 Molecular forces

Fig. 1.1. A: Poisson vs. Laplace. B: schematic representation of hydration, or surface induced liquid structure. Around an interacting molecule (such as an ion) the solvent has to be perturbed by the solute molecules.

surface-induced profile of order? That is, does the liquid density gradually change from the bulk value as we approach the surface (the Poisson view)?

Alternatively, and another statement of the problem: consider two inert solute atoms, molecules or ions dissolved in a liquid, approaching each other under the influence of the same forces. In our modelling, can we consider them as hard spheres interacting across the bulk continuum liquid (Fig. 1.1) (the so-called primitive model)? Or must we recognize that the structure of the liquid that surrounds the interacting molecules is itself perturbed by the molecules (the civilized model)? If the liquid is water, we term the result of these indirect forces hydration forces. Of course hydration and hydration forces exist. One has simply to put different salt crystals in water and measure the change in volume of the solution to see bulk manifestation of hydration effects.

Challis invented a new term, mathematical physics, for what we now call colloid and surface chemistry. He suggested first that measurement of molecular forces might best be accomplished by using the then newly possible interferometric techniques.

But it had to wait 150 years for the experiments of Tabor, Winterton and Israelachvili that first measured the molecular forces between surfaces to confirm the quantum mechanical theories that had emerged in the interim [2,3].
It is salutary to reflect on such matters. Because the fact remains that despite all the advances in experimental techniques that are used by biologists, organic chemists and biochemists, all the progress that has been made in the physical chemistry of liquids over the last 50 years since the molecular biology revolution has contributed very little at a conceptual level to the progress of modern molecular biology.

This is genuinely puzzling to physicists and physical chemists who believe, like D’Arcy Thompson, in reductionism. That is, there ought to be useful connections, at least intersections, between the physical and biological sciences beyond the chanting of ‘non-equilibrium thermodynamics’ or ‘chaos’ in a mantra that becomes dreary.

Is this so, and why is it so? The problem, this lack of confidence in science, is not peculiar to physical chemistry. It is embodied in the ‘science is dead’ bleat of the 1990s.

It is embodied in Morris Klein’s marvellous book, *Mathematics, the Loss of Certainty*. The mathematicians lost their faith after Godel’s theorem proved there was no such thing as absolute proof. There is now not one but many mathematics, little of which is so integrally associated with science as it used to be.

The physicists, with the triumphs of nuclear physics and quantum mechanics and later the solid state, had no doubts about the future and their central role therein. Schrödinger enigmatically speculated somewhat in *What is Life*, and in *Science and Determinism*. Delbruck, a physicist, confronted biology directly with his work on bacteriophages and enthused a generation. Excepting a few hardy biophysicists, the physicists and physical chemists had given up on biology. Some retreated into mumbling on the mystiques attending non-linearity as the source of the New Jerusalem.

1.2 Liquids and computer simulation

A new generation seeks salvation and insights via computer simulations, of liquids, interfaces, membranes and proteins. Perhaps that is as it should be. But nagging doubts remain. The simulation of the structure of a protein in agreement with its X-ray crystal form is a technical triumph. The determination takes up to 30000 individual effective molecular force parameters, parameters that depend on temperature. Something, apart from water, seems to be missing in this kind of modelling. The structure tells us little of function.

The attractiveness of simulation, a new kind of experimental technique, derives in part from the awkward fact that there is no real molecular theory even of simple liquids. This is because, unlike a solid or gas, a liquid has no ideal molecular reference state. For a solid this is a periodic delta function that describes an array
of atoms in a crystal. For a gas a statistical description starts with a completely random distribution of its molecules. This central difficulty for the theory of the liquid state of matter remains.

It is worse for water, the stuff of life. Bernal and Langmuir were probably closer to the mark with their views that water was more like a giant dynamic cooperative entity, as for proteins, rather than a collection of individual molecules. Onsager knew and said the problem was the old one of water. That is indeed a problem.

1.3 Interfaces and colloids

But it is not the only difficulty: we can observe that an interface is a physical reality. A lipid membrane and a biological cell surface exist. In a broader context it is an idealization. The context is in consideration of the totality of real states of matter. Indeed, a macroscopic continuum itself is an idealized notion. Homogeneous gases, liquids and solids that we deal with by thermodynamics never occur other than as theoretical constructs of infinite extent. Soils, clouds, granites and living organisms all are real objects, non-uniform at different levels of organization, even fractal on the entire range of scales, from the atomic to the macroscopic domain.

The usual models of gases or solutions or real crystals are limited in this context. Just as the theory of dislocations in solids allows for some disorder in an ideal array of atoms, so too the virial expansion makes a second-order correction to the ideal gas/solution equation of state. As perturbation theories, these help to account for small deviations from ideality (e.g. the van der Waals equation of state or Debye-Hückel theory of electrolytes). But in passing from the limit of infinite dilution, the mathematical difficulties that confront exact description increase enormously. Additional parameters are invoked to accommodate deviations from limiting laws, and associated uncertainties begin to increase. Theory already breaks down at higher concentrations, even when a gas or a solution still remains homogeneous. It fails and becomes invalid long before the assumption of homogeneity becomes violated by liquid–gas nucleation at supersaturation. Nonhomogeneous and supramolecular systems can hardly ever be explained with standard theories. It is these systems for which a spectrum of ‘unusual’ mechanical and optical properties occurs. These states of matter are excluded from consideration in ordinary solid-state physics and fluid mechanics.

The innovative terms ‘soft condensed matter’ or nanomaterials have come into vogue and have subsumed the old-fashioned term ‘colloids’. But colloids remain despite a relabelling. They are springy liquids and sticky solids that never conform with one or the other of the three states of matter. They do not conform to the structures imposed by the universal triad: gas, liquid, solid.
Reasons for the enquiry

Colloid science attempts to handle the challenge. It has as its aim a theory of all these nonconformist states of matter.

It begins with an unravelling of the problem at its most fundamental level – starting with an interface. Here discontinuity of macroscopic physical properties occurs in its most severe form. The homogeneity assumption here breaks down completely. This seemingly leaves no hope that thermodynamic principles might apply. Equilibrium thermodynamic notions make sense only for uniform continua. Nevertheless, exact thermodynamic results derived for bulk phases do carry over to interfaces. This is because surfaces, although extremely non-uniform, are non-uniform only in a single – normal – direction. In the other two lateral dimensions macroscopic averaging can be done.

Gradients of density and of other thermodynamic properties that occur in a direction perpendicular to a surface are enormous. The associated tangential stress at the surface of a liquid remains at the ultimate limit that a condensed phase is able to sustain. This is the essence of the mechanism by which a liquid terminates at its boundary where it breaks into the vapour. Surface tension is the manifestation of this phase collapse effect. (It is the integral of the profile of this excess pressure that acts in the lateral direction that defines surface tension.)

So interfaces can be investigated by thermodynamics. The theory of capillarity is indeed at the same level of physical rigour that applies to bulk vapours, liquids and solutions. By consideration of different interfaces a great variety of non-uniform chemical compositions that occur in practice can be embraced by a unified physical model.

It is for this reason that thermodynamic quantification of interfaces is such an important issue. By experimenting with interfaces and with their interactions we can extend our exact knowledge further, and then the more complicated systems that we call colloidal dispersions come within reach. The thermodynamic state of a disperse system for which one phase is finely distributed in another is fully defined in terms of interfacial and bulk phase properties.

While a colloidal particle may be compositionally identical to the bulk phases from which it forms, a colloidal dispersion is altogether a quite different entity. It is entirely different in many respects from its parent macrophases. So, for example, emulsions formed from a mixture of transparent Newtonian liquids are opaque and sometimes solid-like. Chemically reduced down to its metal form, gold in a colloidal sol, due to Brownian motion of colloidal particles and double layer repulsion between their surfaces, remains ‘dissolved’ – suspended in water. The theory of such systems is an extension of molecular-kinetic theory into the world of colloidal dimensions. It is not necessary to give formal mathematical descriptions to conclude that all the amazing diversity of flow and colour patterns seen are brought into existence by interfacial gradients of thermodynamic properties. These occur over molecular distance scales.
1.4 Colloids, polymers and living matter

The word colloid is a term originally associated with dairy processing and similar biotechnologies. The very term coagulation is a notion derived from observations on the clotting of blood.

Colloidal states of matter were first taken to be a specific property of polymers. These giant molecules, far in excess of ordinary molecular sizes, were believed to occur in living matter. Many of these components of bioextracts, of which common examples are soaps or gelatine, have been known since ancient times. They were isolated and identified chemically following progress in biochemistry, often as individual compounds, in the eighteenth and nineteenth centuries. They are not always of very high molecular weight. These molecules nevertheless avoid the formation of ordinary crystals and solutions. They prefer rather to persist in their own micellar and liquid crystalline and amorphous states. A jellyfish with up to 98% water is a challenging example.

It was not until the turn of the twentieth century that it was realized that colloid behaviour is not a unique property of biomaterials. It extends into the realm of inorganic chemistry as well. Swollen clays in soils and gold hydrosols are examples.

1.5 Conceptual locks

There was much progress in colloid and polymer science in the second half of the twentieth century. Rather there appeared to be progress. We shall come to this in detail later.

But the nagging problem of intersections, communication between the physical and biological sciences remained. All the progress in the physical sciences still seemed irrelevant, in this sense: biologists and biochemists certainly use the tools developed by physical chemistry and other physical sciences. They use pH meters, buffers, electron microscopes and nuclear magnetic resonance to measure ‘ion binding’ of ions to proteins. They use X-rays to determine protein structure, ultracentrifuges to sort macromolecules. Electrophysiologists use techniques of electrochemistry to measure membrane potentials. But at a conceptual level there is a disjunction.

1.6 The nub of the matter

In seeking an insight into our problem, we take a remark of Stephen J. Gould, who in one of his books on evolution, Eight Little Piggies, wrote: ‘I have long maintained that conceptual locks are a far more important barrier to progress in science than factual lacks’. In that aphorism lies a clue to our dilemma. If we can identify those conceptual locks, we might hope to make progress.
To make our case we have first to agree on what constitutes a theory, and then to identify the barriers to progress. The word ‘definition’ has defied definition by the philosophers. We can probably agree, however, that the first stage of science is the naming of things, whence follows awareness of similarities and relations between them. Once those connections can be ordered into a predictive dictionary of events, we have a real theory. The fewer the parameters required to accomplish the ordering, the better is the theory.

For example, the Ptolemaic theory of planetary motion worked quite well. But the Newtonian theory is better, and predicts more. The conceptual lock here, which required more and more parameters as time went on, was evidently the notion that the earth is the centre of the universe. Another example is the modelling of a liquid via computer simulation. With the equations of statistical mechanics only about eight parameters, to specify a molecular potential, are required, to account for the phase diagram of a simple liquid such as argon, given sufficient computer time. But the success of such a computation is tempered a little by the realization that one might do just as well with the same number of parameters, and with molecular potentials of a completely different functional form that have no connection with the actual potentials.

We can agree that thermodynamics works. It must, being a tautology. So instead of doing the statistical mechanics of interacting molecules via simulation, one might just as well measure the boiling or freezing point of argon if one wants those properties.

This trivial example is not so far removed from our theme as first appears. As already remarked, the use of an army of molecular parameters to simulate the folding of proteins is currently popular. Yet from another point of view it is Ptolemy gone mad. Insight is obscured. To simulate what happens when the protein denatures, say, over a very narrow temperature range, one simply changes the parameters. The same used to be true even for such a deceptively simple problem as the calculation of the activity coefficients and osmotic pressure of mixed electrolytes. Change the mix. Then change the parameters. These preoccupations of physical chemists are not theories that will be useful to biologists, or chemical engineers. In this example, the sole point of a theory of activity coefficients or osmotic pressure of simple electrolytes is to see whether the primitive model (continuum solvent approximation) is a valid approximation. It is, and that is useful, for 1:1 alkali halide salts, at low concentrations where the ionic interactions are very long-range. The model fails for interesting ions such as sulfate, nitrate, caesium, phosphate, and for all of them at the concentrations of interest to biology. If we cannot predict the osmotic pressure of a sodium chloride solution in water we are going to be in trouble with a solution of proteins.
1.7 Molecular forces in self assembly

The conceptual lock here turns out to be in part due to the too-simple approximation that the solvent, water, is a continuum. But it is due too to the fact that all the classical models of electrolyte chemistry depend on electrostatic models of ionic interactions. These ignore all the quantum mechanical forces acting between ions that are just as important, and determine the very specificity that we are looking for!1

1.7 Molecular forces in self assembly

If we come to self-assembling amphoteric molecules like the phospholipids of cell membranes we enter into more difficult ground. Here is a statement of our problem that sums it up: ‘Despite enormous progress in understanding the genetics and biochemistry of molecular synthesis we still have primitive ideas of how linearly synthesized molecules form the multimolecular aggregates that are cellular structures. We assume that the physical forces acting between aggregates of molecules and between individual molecules should explain many of their associative properties; but available physical methods have been inadequate for measuring these forces in solids or liquids.’ These few succinct opening sentences from an old review by V. A. Parsegian stand [5]. They embrace and define the whole grey area bridging chemistry, physics and biology which is our concern. They imply a formidable injunction. For while it is axiomatic to the physicist or chemist that structural changes in any system should be reduced to a consideration of forces or free energies which cause those changes, the burden of proof lies with the proponent. The axioms of physics do not always receive so ready an acceptance from biologists whose whole thinking in the past has been centred on the role of geometry to the almost complete exclusion of forces and entropy. The burden of proof becomes especially great if one considers the increasing sophistication of those few successful theoretical advances in our understanding of condensed matter. To be convincing, and to have any hope whatever of reducing to some semblance of order the vast complexity of those intricate multimolecular structures that are the subject of biology, any successful theories must have as a minimal requirement extreme simplicity, to make them accessible to the biologist who has enough concerns of his own not to be dragged into the subtleties of modern physics.

1 An extreme example of the absurdities this path to theory can lead to is the idea that if one put all the nuclear particles and electrons together and wrote down their potentials of interaction and solved the equations one would come up with a universe that is us. It works, say for the excitation spectra of complicated molecules, but in the process all notions of chirality are lost. The Born–Oppenheimer approximation that fixes the positions of the nuclei does retain shape, but at the expense of a loss of computational accuracy.