Complex carbohydrates such as the high molecular weight polysaccharides starch and cellulose are well known for their importance in foodstuffs, paper and wood, but their importance extends far beyond that into the biopharmaceutical, healthcare, oil and printing industries. In many cases their ability to be degraded is highly favourable – for example in nutrition, paper/agro-waste, recyclisation and their use as potential biofuels. In other instances retarding degradation is preferred – in their use as biopharmaceuticals for example, or in preserving old wood structures in archaeology. A Special one-day Discussion Meeting, jointly run by the RSC Biotechnology & Carbohydrate Groups – which also formed a Satellite of the European Polysaccharide Network of Excellence (EPNOE) meeting that had been held in Wageningen the week previous – was therefore put together to bring together representatives from the polysaccharide and glycoconjugate communities to review and discuss in detail one specific area of carbohydrate polymer research, namely the importance of the stability and degradation of these substances – the biochemical mechanisms involved and methods for studying stability and degradation at the molecular level.

The meeting began with Professor Felix Franks (Bioupdate Foundation, London) who gave a talk entitled Carbohydrates: First cousins of water. In considering the structure of polysaccharides he showed how it was important when considering stability issues just how close to water these “polyhydrate” molecules are. He considered the most important resemblance between water and carbohydrates is their common feature of a lack in the ability to form covalent bonds; all interactions rely on weak hydrogen bonds in which the sp$^3$ oxygen orbital hybridisation dictates the molecular conformation (tetrahedral). Water as an amphiphatic molecule, can participate in a wide range of reactions – proton donor/acceptor, oxidation/reduction, and hydrolysis/aggregation – and to some extent carbohydrates and polysaccharides are able to participate in the same types of reactions, although reaction rates in fused or vitreous materials will differ vastly from aqueous solution. Those are properties which Greg Tucker (University of Nottingham) followed up with in his presentation on the Enzymatic degradation of cell wall polysaccharides in which he described the many advances in our knowledge of the action and genetics of pectinesterase enzymes in fruit. Anna Suurnäkki (VTT Technical Research Centre of Finland and EPNOE) then described in her talk on Enzymatic treatment of polysaccharides how the susceptibility of polysaccharides to enzyme modification could be put to good use in various applications. She explained how the three main enzyme-catalysed reactions of polysaccharides are hydrolysis, oxidation and transfer reactions. These reactions can result in partial or total depolymerisation, specific oxidation and group transfer reactions of polysaccharides, respectively. Enzymes can thus be exploited in both degradation and targeted modification of polysaccharide structure. The right selection of enzyme, reaction conditions and process optimisation is, however, needed to achieve a cost-efficient, industrially feasible enzymatic process or process stage, and a particularly good example of the
potential of enzymes in modification and processing was in the application to lignocellulosics and their main polysaccharides, cellulose and hemicellulose.

Peter Ellis (Kings College London) then described in his talk *Dietary Fibre: Functional components and mechanisms of action in the gastrointestinal tract* how the rate and extent of starch and lipid digestion in the gut could be strongly affected by edible plant polysaccharides, notably the water-soluble viscosity-producing polymers such as legume galactomannans. Moreover, studies of almond seeds have shown that the physical state of dietary fibre is crucial, so that intact cell walls post-ingestion can encapsulate lipids and therefore hinder the digestion process. Thus only lipid from ruptured cells following mastication is bioaccessible and available for digestion. Predictions of lipid bioaccessibility, estimated from a theoretical model he presented, appear to agree reasonably well with the empirical data obtained from digestibility studies *in vitro*. However, the *in vivo* data indicated that lipid is also released from intact almond cells, but at a much slower rate (>3 h post-ingestion). Thus, overall a biphasic mechanism for lipid release appears to be operating. The theoretical model has the potential to be developed further to study lipid bioaccessibility in almonds (used as a model system) and also to study the bioaccessibility of other nutrients and plant tissues.

Bioencapsulation was also the subject of the presentation by Christine Wandrey (Ecole Polytechnique Fédérale de Lausanne) entitled *Stability of polysaccharide encapsulation complexes*. She explained the importance of hydrophilic nanocarriers as vectors in biomedical and pharmaceutical applications, particularly those involving complexes of chitosan with alginate. She showed how hydrodynamic methods (dynamic light scattering, zeta potential and analytical ultracentrifugation) delivered reliable characteristics with a clear dependence on ionic strength and temperature (as expected). Also strong deformability and stability in different media have been reported as crucial parameters, and this appeared to be the case if either fungal or animal sources of the chitosan prior to linkage with tri-polyphosphate (TPP) are involved.

The meeting then moved from polysaccharide assemblies to include the stability of glycoconjugates with a presentation by Chris Jones (National Institute of Biological Standards) on *Polysaccharide and glycoconjugate vaccines based on bacterial surface glycans*. He showed the contrasting dependencies of the effectiveness of both on molecular weight distribution. Different strategies for glycosylation lead to different stabilities against glycan degradation: physicochemical analysis of the native polysaccharides and final conjugates is providing insights into the mechanism by which they degrade. A further important class of glycoconjugates are the mucin glycoproteins, which form an integral part of the mucosal defensive barrier at surfaces throughout the body. As the barrier is closely integrated with innate and immune defensive systems it has a requirement for controlled turnover while retaining defensive functions on a continuous basis.

Tony Corfield (University of Bristol) described the current understanding of *Mucin turnover* – this is a balance between biosynthesis, oligomerisation and network formation, physical disruption at the mucosal surface and the action of specific enzymes which degrade the glycan chains and the peptide backbone. An important feature of this process is the enzymes which generate and cleave disulphide bridges.
found within the mucin monomers themselves, and also between monomers to yield dimers and oligomers. Evidence suggests that the formation of mucin fragments is a fundamental part of the turnover process and that these are generated on a tissue- and mucin-specific basis.

Steve Harding (National Centre for Macromolecular Hydrodynamics, Nottingham, and EPNOE) then described *Advances in hydrodynamic stability probes* – and in particular a new analytical ultracentrifuge approach to macromolecular stability and illustrated this with application to the determination (without possible artificial features from columns or membrane based methods) of a molecular weight distribution of mucins, glycoconjugate vaccines and polysaccharides. Gordon Morris (University of Huddersfield) then described the application of a variety of hydrodynamic probes to a study of the *Stability of chitosan and pectin as nasal and intestinal mucoadhesives*. Both chitosan and pectin depolymerise after prolonged storage periods (up to 1 year) and the rate of depolymerisation is greater at elevated temperatures (40 °C). This results in the disintegration of TPP-chitosan nanoparticles and a decrease in the strength for LM pectin gels, although this decrease in gel strength does not have a detrimental effect on the release of a model drug.

In the final part of the meeting there then followed a series of papers on cellulose and lignins. Michael Jarvis (University of Glasgow) in his presentation *Cellulose crystallinity: perspectives from spectroscopy and diffraction* described how, because of the stability of crystalline cellulose, the degree of crystallinity has a large effect on rates of cellulose degradation in almost every situation where cellulose is degraded, and how X-ray diffraction, solid-state NMR and FTIR spectroscopy and neutron scattering are giving us powerful insights into this stability and in particular the role of the interplay between water immobilisation and microfibril structure. Patrick Navard (Centre de Mise en Forme des Matériaux at Sophia Antipolis and EPNOE) in his talk entitled *Cellulose de-construction: what can be learned from molecular modeling and dissolution experiments?* showed that to understand degradation and stability it was important to understand the complex way in which cellulose chains are synthesized and crystalline cellulose is formed - and the important correlation between the stresses on cellulose chains and dissolution and stability. When cellulose chains leave a cell membrane they form a disordered pile on the surface before being pulled out by cellulose synthase activity.

Callum Hill (Edinburgh Napier University) in his talk *Water vapour sorption properties of cellulose – the parallel exponential kinetics of model and cell wall viscoelasticity* then showed the importance of moisture content on stability of cellulose and lignin structures in terms of the parallel exponential kinetics (PEK) model using Kelvin-Voigt based considerations. In the case of a cellulosic fibre subjected to a change in relative humidity, there is a change in the swelling pressure exerted within the cell wall when the atmospheric water vapour pressure is raised. Then Claire Halpin (University of Dundee) described how *Lignin* is a major barrier to the efficient conversion of complex wall polysaccharides (cellulose and hemicellulose) into the simple sugars that could be used as substrates for liquid biofuel and speciality chemical production. The lignin content and composition can be manipulated at a genetic level by altering the expression of lignin biosynthesis genes and without - depending on the gene being targeted - impacting on plant health and fitness, even in field-grown plants. An
understanding of these processes is providing valuable information that can be used to devise effective strategies for the future breeding of plant biomass improved for bioenergy applications.

The final presentation considered the stability of archaeological wood structures. Nanna Bjerregaard Pedersen (University of Copenhagen) in her presentation on *Bacterial degradation of historical wood timbers found in near anoxic waterlogged environments* showed how all this increased knowledge at the molecular and microbiological level is proving crucial to our understanding of the stability and the assessment of decay profiles of buried ancient shipwrecks. Historical wood timbers can survive for hundreds or thousands of years in anoxic waterlogged environments due to the lack of oxygen supply for the aggressive wood degrading fungi, wood boring insects and wood boring molluscs. Erosion bacteria cause decay of the secondary cell wall in anoxic environments. New studies with cellular UV-microspectrophotometry (UMSP) of decayed tracheids of spruce and pine wood have shown that residual material left from erosion bacterial decay contains lignin. In addition the analysis show a so far unknown decrease in lignin content in the tangential middle lamella. The Comprehensive Microarray Polymer Profiling (CoMPP) technique - for characterizing plant cell wall carbohydrates - can also be used to gain a greater knowledge of which carbohydrate structures are preferentially decayed by erosion bacteria.

The meeting - which was oversubscribed – was held in the Council Room at the historic Burlington House, home of the Royal Society of Chemistry in London and was generously sponsored by the Chemistry Biology Interface Division of the RSC together with Nestle (York) and Glycomix Ltd (Reading). This support enabled costs to be kept down particularly for the many younger scientists who participated: feedback from them said how much they had thoroughly enjoyed and benefited from being in the presence of distinguished experts in their respective fields. Many of them presented posters during the extended lunch and tea breaks at the meeting – all were of very high quality - and the prize of £100 for the best poster was awarded to Dr. Terri Grassby (Kings College London) for her poster on *Plant cell walls as barriers to lipid bioaccessibility in a model plant food: in silico methods for estimating lipid release.*

Poster prize winners: *(left to right)* Kenzi Clark (Institute of Food Research, Norwich) and Fuad Hajji (Nottingham) (joint 2nd prize) with Terri Grassby (Kings College London), winner of the 1st Prize. Far right is Prof Rob Field (Chairman, RSC Carbohydrate Group).