

Summary

The form of organisation of amphiphilic compounds is of great importance for several biological and technical applications. Nevertheless the structure-property relationship of these compounds has in many areas not been investigated thoroughly yet. Especially the influence of chirality and complex structures is in many parts still not clear, in particular regarding the difference in energetically comparable structural order. The aim of this work is largely the investigation of the form of organisation of micelles, vesicles and membranes in aqueous media, as well as their interaction and transitions.

Different structures from simple alkyl glycosides, alkyl glycosides with spacers to complex glyco glycerolipids can serve as target compounds for these investigations.

The investigation of these properties requires the use of different characterisation methods and their combinations, as well as theoretical concepts.

Liquid crystalline properties were determined using polarising microscopy. The thermotropic phase behaviour was determined by heating the sample, whereas the lyotropic phase behaviour it is determined by the contact of the sample with water. In case of the lyotropic phase behaviour it is possible to observe the complete phase sequence over the whole concentration range. The characterisation of the single phases requires different physical methods. Information about the calorimetric data of the phase transitions determined by differential scanning calorimetry (DSC) accomplish the results from the polarising microscopy, but is restricted to the investigation of defined concentrations.

The aggregation of amphiphiles in solution starts above the critical micelles concentration (*CMC*). The *CMC* was determined from the change of the surface tension in dependence of the concentration of amphiphile in solution, using a tensiometer. Furthermore it is possible to determine the area of an amphiphile molecule at the air-water interface, as well as to predict the possible shape of the micelles formed above the *CMC*.

The aggregate structure of amphiphiles in dilute solution (< 5 wt%) was determined using small-angle neutron scattering (SANS). The micellar form and structure can be determined very exactly due to the different scattering behaviour of neutrons on the nuclei of hydrogen and deuterium. In some cases it was also possible to determine the change of the micellar structure, in dependence of the concentration or temperature. The micellar structures were calculated using two different ways, a model-independent approach and fitting of the scattering data to different models. Comparing the results of both approaches gives concise information on the studied systems.

For the characterisation of the higher ordered lyotropic phases, especially of the biological relevant lamellar and cubic phases, small-angle x-ray scattering (SAXS) experiments were performed. Additional information about the packing order of the hydrophobic alkyl- and acyl chains was retrieved from the investigation with fourier-transform-infrared-spectroscopy (FTIR).

The foaming properties of amphiphiles are an important technical feature of these compounds. The foaming behaviour was determined using a modification of the standard procedure according to DIN 53902-2, which gave reliable results, but required only very small amounts of amphiphile. The in-vivo characterisation of the foam structure and aging of the foams was made using a modified light microscope. These gave insights into the foam structure and the statistic pore size distribution of the air-bubbles inside of the foam.

The theoretical concept of the hydrophilic-lipophilic balance (*HLBG*) was accomplished by practical tests. The concept of the critical packing parameter (*CPP*), which predicts the possible aggregate form in dilute solution, was compared with the results from the SANS-experiments.

The investigated compounds were synthesised using several synthetic pathways, which gave the target compounds in gram scale and in highest purity. The different structural types covered ranged from alkyl glycosides and alkyl glycosides with different types of spacers to complex glyco glycerolipids. The molecular shape was also varied by the use of different carbohydrates (Glucose, Lactose, Maltose, Melibiose, Gentiobiose, Maltotriose). For some compounds the synthetic pathways were optimised, which gave the compounds in considerable higher yields and, in part, less synthetic

steps. It was also observed, that the differences in the physical properties of glyco glycerolipids with the alkyl chain connected via an ether or ester linkage are very small. Moreover the 1,3-ether lipids can be synthesised in a few steps in gram scale, whereas the synthesis of the ester based lipids requires numerous steps. From 49 lipids synthesised in this work, 32 were synthesised for the first time.

The thermotropic liquid crystalline properties of the investigated alkyl glycosides differ mainly in the transition temperatures. High melting points were determined for compounds with linear alkyl chains. The melting point is lower for compounds with unsaturated and with methyl branched alkyl chains compared to the corresponding analogues with linear alkyl chains. In this case most of the compounds are in the liquid crystalline state at room temperature. Exceptions were found for compounds with unsaturated alkyl chains containing amide groups in the hydrophilic or in the spacer part. Here the melting point is above room temperature. Nearly all alkyl glycosides with disaccharide headgroups formed exclusively a smectic A phase. Only for three compounds was a cubic phase observed on cooling.

Glyco glycerolipids with trisaccharide carbohydrate headgroups or with two methyl branched alkyl chains only show smectic A phases, too. Interestingly it was found that 1,3-di-*O*-alkyl-glycerolipids show nearly the same phase behaviour and transition temperatures as the corresponding glycosyl diacyl glycerols. The formation of columnar phases besides the smectic A phase was observed for slightly wedge shaped compounds, whereas Y-shaped glyco glycerolipids and lipids with tilted headgroups also have the ability to form cubic phases.

Conversely to thermotropism, the lyotropic phase behaviour is already affected by small changes of the molecular structure. Generally it was found that, with the exception of Y-shaped glyco glycerolipids, all compounds investigated in this work show lamellar phases and so have the ability to act as membrane forming or membrane stabilising compounds. In the case of glyco glycerolipids with disaccharide headgroups and linear alkyl chains inverted phases are formed, whereas most of the alkyl glycosides form hexagonal and partly lyotropic cubic phases. An interesting exception was found for compounds with an ethanolamine spacer. Except for one compound it was observed that the spacer allows only the formation of lamellar phases. Investigation of the lyotropic aggregate structure of the lipid 1,3-di-*O*-(*cis*-9-octadecenyl)-2-*O*- β -D-maltopyranosyl-sn-glycerol showed the formation of a well-resolved cubic phase over a broad concentration and temperature range. This result is of high biological relevance as such behaviour has so far only been observed for a few lecithine lipids.

Y-mesogenes exhibit thermotropic and lyotropic cubic phases. The structure enables the formation of cubic structures. Supposedly this structure facilitates the formation of cubic phases in biological processes.

The chemical structure of the amphiphiles strongly influences the *CMC*. Additionally it was found that amphiphiles with a Maltose carbohydrate headgroup are more surface-active than amphiphiles bearing Melibiose or Maltotriose headgroups.

Micelle formation was observed for several amphiphilic lipids. A more detailed concept for the formation of micelles by carbohydrate-based amphiphiles could be derived from this data. The change of the micellar structure can be explained using a general structural concept. Compounds of a rod-like molecular shape with *n*-alkyl chains form large aggregates. Amphiphiles with a wedge-shaped or tilted structure and a *n*-alkyl chain show a critical chain length above which rod-like micelles are formed and below it spherical micelles. The growth of rod-like micelles of the investigated lipids was determined to follow standard models. Spacers will change the micellar structure. Ethylene glycol spacers increase the hydrophilic part of the molecule and also the flexibility of the alkyl chain. Ethanolamine spacers connected with the fatty acid chain by an amide group also increase the hydrophilic part, but the flexibility is dramatically reduced. In contrast an ethyl branching in the spacer part of ethanolamine increases the hydrophobic part.

Short methyl branched chains, instead of *n*-alkyl chains, favour the formation of bilayer structures whereas long methyl branched chains will form micellar structures. Large carbohydrate headgroups (trisaccharides) as well as broad hydrophilic headgroups (Y-shaped lipids) can only form spherical

micelles. Also bolaamphiphiles with two disaccharide headgroups can form spherical micelles, but only in a very narrow concentration range, most probably followed by the formation of bilayer structures. This effect shows, that the interaction between membranes formed by the bolaamphiphile and vesicles or micelles is different as it is for non-bola lipids

The interaction of a carbohydrate-based bolaamphiphile with octyl β -D-glucopyranoside was found to be antagonistic, which is confirmed by a higher mixture *CMC* compared to the ideal mixing expectation and a decreasing of length of octyl β -D-glucopyranoside micelles upon addition of a small amount of bolaamphiphile to the solution. These results are unexpected, because the chemical structures of the investigated compounds are very similar and thus an increase in the size of micelles would normally be expected.

Thermotropic properties can be related to the micellar structure in dilute solution. Compounds showing a complex thermotropic phase behaviour due to a labile balance between the hydrophilic and hydrophobic moiety form large aggregate structures already at low concentrations in aqueous solution. A simple thermotropic phase behaviour is connected with the formation of small micelles in dilute solution.

Prediction of the aggregate form above the critical micelle concentration (*CMC*) by the Critical Packing Parameter (*CPP*), derived from the headgroup area at the air-water interface is only of limited use for carbohydrate-based amphiphiles. The micelle shape moreover depends on small changes of the molecular structure not captured by the adsorption at the liquid-air interface.

The relationship between the chemical structure and the micellar forms can be used for the design of micellar solutions in new applications.

The aqueous solubility of the amphiphiles in this thesis follows the general trend expected from their hydrophilic-lipophilic balance according to Griffin (*HLBG*), e.g. good water solubility was found for amphiphiles with a high *HLBG* number and a good emulsification ability was found for lipids with low *HLBG* number.

The method of methylation of the alkyl chain controls the self-assembly and can explain different biological functions for either plants (variable temperature) or animals (constant temperature). The solubility of the melibioside (2*R*,4*R*,6*R*,8*R*)-2,4,6,8-tetramethyldecyl-6-*O*-(α -D-galactopyranosyl)- β -D-glucopyranoside (MeI- β -TMD) was found to be low even above its Krafft-temperature. This is similar to observations made for the (all-*R*) tetramethyldecyl wax esters of the preen gland, which is responsible for the good water protection of the feathers of the domestic goose. The TMD-chains retains this effect even as a part of a more hydrophilic molecule. Additionally a methyl-branched chain favours the formation of membranes. Conversely the formation of cubic and columnar phases is suppressed.

Carbohydrate based amphiphiles are good foaming agents above their Krafft-temperature already at very low concentrations. Foams formed by the investigated compounds are very stable, especially compared to technical surfactants. The foaming ability increases with increasing temperature. The results give new insights in the relationship between the chemical structure of the amphiphiles and the foaming behaviour, which allows the selection of an appreciable foamer. For use as cosmetic detergent it was found, by foaming microscopy, that these new amphiphilic sugar surfactants have the ability to form very fine structured foams, with a very narrow pore-size distribution of small air-bubbles. Also the stability of the foam is superior compared to standard cosmetic surfactants.

The general concepts, derived from the phase behaviour of the investigated simple lipid structures can be validated in future for its use on more complex and natural structures.