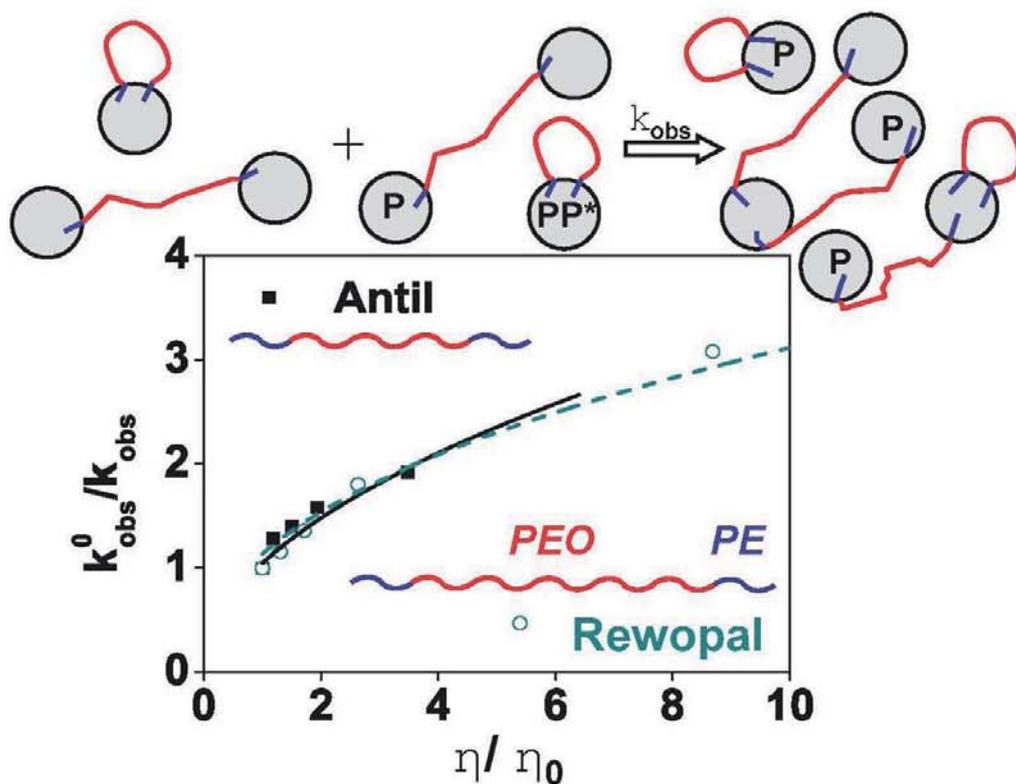


---

## Dynamics of Solubilisation Processes in Amphiphilic Systems Studied by Highly Time-Resolved Stopped-Flow Experiments

---





# **Dynamics of Solubilisation Processes in Amphiphilic Systems Studied by Highly Time-Resolved Stopped-Flow Experiments**

vorgelegt von  
Diplom-Chemikerin  
**Anina Barth**  
Potsdam

Von der Fakultät II - Mathematik und Naturwissenschaften  
an der Technischen Universität Berlin  
zur Erlangung des akademischen Grades

Doktor der Naturwissenschaften  
(Dr. rer. nat.)

genehmigte Dissertation

Promotionsausschuss:

Vorsitzender:	Prof. Dr. Reinhard Schomäcker
1. Bericht:	Prof. Dr. Michael Gradzielski
2. Bericht:	Prof. Dr. Joachim Koetz

Tag der wissenschaftlichen Aussprache: 23.08.2011

Berlin 2011  
**D 83**

**Bibliografische Information der Deutschen Nationalbibliothek**

Die Deutsche Nationalbibliothek verzeichnet diese Publikation in der Deutschen Nationalbibliografie; detaillierte bibliografische Daten sind im Internet über <http://dnb.d-nb.de> abrufbar.

1. Aufl. - Göttingen : Cuvillier, 2011

Zugl.: Berlin, Univ., Diss., 2011

978-3-86955-974-5

© CUVILLIER VERLAG, Göttingen 2011

Nonnenstieg 8, 37075 Göttingen

Telefon: 0551-54724-0

Telefax: 0551-54724-21

[www.cuvillier.de](http://www.cuvillier.de)

Alle Rechte vorbehalten. Ohne ausdrückliche Genehmigung des Verlages ist es nicht gestattet, das Buch oder Teile daraus auf fotomechanischem Weg (Fotokopie, Mikrokopie) zu vervielfältigen.

1. Auflage, 2011

Gedruckt auf säurefreiem Papier

978-3-86955-874-5

## Abstract

In this work a comprehensive study of the solubilization dynamics in ternary and quaternary surfactant systems is presented. Microemulsion either with the zwitterionic tetradecyldimethylamine oxide or the nonionic pentaethyleneglycol monododecyl ether were employed to study intensively the dynamics of the exchange of oil between microemulsion droplets. The stopped-flow method - in which two volumes of solutions are rapidly mixed and the properties of the mixed solution followed as a function of time and - was employed intensively throughout this work. Complementary stopped-flow measurements (turbidity, fluorescence and Small Angle X-ray Scattering) served as a suitable tool to monitor the kinetics of solubilisate exchange between and the equilibration of microemulsions and in addition the process of formation of vesicles. For that purpose pyrene butyric acid stearyl ester was specially designed and used as fluorescent probe and by a systematic variation of the molecular composition of the microemulsion a comprehensive understanding of the solubilization dynamics has been achieved.

Fluorescence measurements proved that not the bending elasticity but the curvature of the surfactant monolayer is the decisive factor during the exchange of oil in a ternary, nonionic microemulsion system. Consequently, at identical values of the bending elasticity, coalescence is slower the smaller the aggregates. Furthermore, through addition of small amounts of charge (introduced by admixing an ionic surfactant) and amphiphilic polymers, the coalescence of microemulsions necessary during the process of oil exchange and equilibration, could be decelerated significantly due to the electrostatic and steric repulsion induced in the system. At the same time the activation energy for these two processes was found to increase as a consequence of the hindrance to coalescence.

In a second approach the structural transition from micelle/microemulsion to vesicles was investigated. The spontaneous formation of the vesicles depends strongly on the preparation of the sample and are therefore controlled by the kinetics of their formation process and was found to be slower the higher the distance between the starting composition and the final composition.

# Zusammenfassung

Die vorliegende Arbeit beschäftigt sich mit der Solubilisationsdynamik in ternären und quaternären Tensidsystemen. Es wurden Mikroemulsionen, bestehend aus dem zwitterionischen Tetradecyldimethylaminoxid oder dem nichtionischen Pentaethylglycolmonododecylether, verwendet, um die Ölaustauschkinetik zwischen diesen Mikroemulsionstropfen zu untersuchen. Die Stopped-flow-Methode – bei der identische Volumina zweier Lösungen sehr schnell gemischt werden und die Eigenschaften der Mischung als Funktion der Zeit untersucht werden können – wurde für diese Untersuchungen eingesetzt. Komplementäre Detektionsmethoden (Trübung, Fluoreszenz und Kleinwinkelröntgenstreuung) dienten dazu, die Kinetik des Ölaustausches und der Bildung von amphiphilen Strukturen zu untersuchen. Zu diesem Zweck wurde ein Pyrenstearylester synthetisiert und als Fluoreszenzfarbstoff verwendet, der es ermöglichte, einen umfassenden Überblick über die Solubilisationskinetik zu erlangen, während die molekulare Zusammensetzung der Mikroemulsionen systematisch verändert wurde.

Fluoreszenzmessungen zeigten, dass nicht die Biegesteifigkeit, sondern die Krümmung der Tensidschicht der entscheidende Faktor des Ölaustauschs in einem ternären, nichtionischen Mikroemulsionssystem ist. Bei gleicher Biegesteifigkeit, verläuft der Ölaustausch umso langsamer, desto kleiner die Aggregate. Durch Zugabe kleiner Mengen von Ladung und amphiphilen Polymers und der daraus resultierenden zunehmenden elektrostatischen und sterischen Abstoßung, wurde die Geschwindigkeit des Koaleszenzprozesses gezielt verlangsamt. Gleichzeitig stieg die Aktivierungsenergie für diesen Schritt aufgrund der zusätzlichen Behinderung an.

Ein weiteres Ziel war die Untersuchung des Strukturübergangs Mizelle/Mikroemulsion zu Vesikeln. Die spontane Bildung der Vesikel wird durch die Zusammensetzung der Probe beeinflusst und hängt daher entscheidend von deren Bildungsprozess ab. Die Bildungsgeschwindigkeit ist dabei umso langsamer, je größer der Unterschied in der Zusammensetzung des Anfangs- und Endzustandes.



# Table of Contents

<b>1 Introduction</b> .....	1
<b>2 Experimental Section</b> .....	15
2.1    Methods.....	15
2.1.1    Stopped-flow method.....	15
2.1.2    Fluorescence spectroscopy.....	22
2.1.3    Scattering methods.....	23
2.1.4    Zero-shear viscosity measurements .....	30
2.1.5    Interfacial tension measurements.....	31
2.1.6    Surface tension measurements .....	31
2.1.7    Conductivity measurements.....	33
2.1.8    Rheology .....	33
2.1.9    NMR-spectroscopy .....	34
2.2    Materials and preparation methods .....	35
<b>3 The kinetics of solubilisate exchange in a nonionic surfactant system - correlation of measured rates with interfacial rigidity, charge and addition of polymer</b> .....	38
3.1    Introduction.....	40
3.2    Results and Discussion .....	45
3.2.1    Influence of chain length of solubilised alkanes and temperature .....	45
3.2.2    Influence of HM-polymer .....	55
3.2.3    Influence of charge .....	57
3.3    Discussion.....	59
3.4    Conclusion .....	63
<b>4 Kinetics of the equilibration of microemulsions and the exchange of oil in a quaternary surfactant system</b> .....	64
4.1    Introduction.....	66
4.2    Experimental .....	68
4.3    Results and Discussion .....	69
4.3.1    Phase behavior .....	69
4.3.2    Kinetics of the exchange of oil between microemulsions .....	70
4.3.3    Kinetics of the equilibration of microemulsions.....	75
4.4    Conclusions.....	83

<b>5</b>	<b>The kinetics of solubilisate exchange in a zwitterionic microemulsion on addition of hydrophobically end-capped poly(ethylene oxide)</b>	<b>85</b>
5.1	Introduction	87
5.2	Results and Discussion	88
5.2.1	Viscosity measurements	88
5.2.2	Kinetic of the exchange of solubilisate	89
5.3	Discussion	93
5.4	Conclusion	97
<b>6</b>	<b>Solubilisation of different medium chain esters in zwitterionic surfactant solutions – effects on phase behavior and structure</b>	<b>98</b>
6.1	Introduction	100
6.2	Experimental	103
6.3	Results and discussion	106
6.3.1	Phase behavior	106
6.3.2	Viscosity	111
6.3.3	Small-angle neutron scattering	112
6.4	Conclusion	121
<b>7</b>	<b>Solubilisation of medium chain esters in zwitterionic surfactant solutions - kinetics of solubilisation</b>	<b>123</b>
7.1	Introduction	125
7.2	Experimental	126
7.3	Results and Discussion	127
7.3.1	Phase behavior	127
7.3.2	Influence of charge	128
7.3.3	Dynamics of spontaneous formation of vesicles – stopped-flow measurements	129
7.3.4	Transmission measurements	130
7.3.5	Conductivity measurements	135
7.3.6	Small angle X-ray measurements	138
7.3.7	Dynamic light scattering measurements	140
7.4	Conclusion	142

<b>8 Conclusion and Outlook</b> .....	144
<b>9 Appendix</b> .....	A
9.1 Appendix to Chapter 3 .....	A
9.2 Appendix to Chapter 4 .....	F
9.3 Appendix to Chapter 5 .....	G
9.4 Appendix to Chapter 6 .....	H
9.5 Appendix to Chapter 7 .....	J

# 1 Introduction

In aqueous solution many surfactants exhibit a rich phase behavior due to their ability to self-assemble into a large variety of morphologically different structures. The simplest ones are micellar structures of e.g. spherical or cylindrical shape to more complex ones like lamellar, vesicle or sponge phases.

Originally proposed by Tanford [1] but later refined, Israelachvili et al. [2] introduced the concept of the so-called packing parameter in 1976, a theoretical framework that relates molecular parameters (head group area, chain length, hydrophobic tail volume) and intensive variables (temperature, ionic strength etc.) to surfactant microstructures and which provides the opportunity to 'tailor' the amphiphilic morphology as desired.

The (critical) packing parameter  $P$  is given by

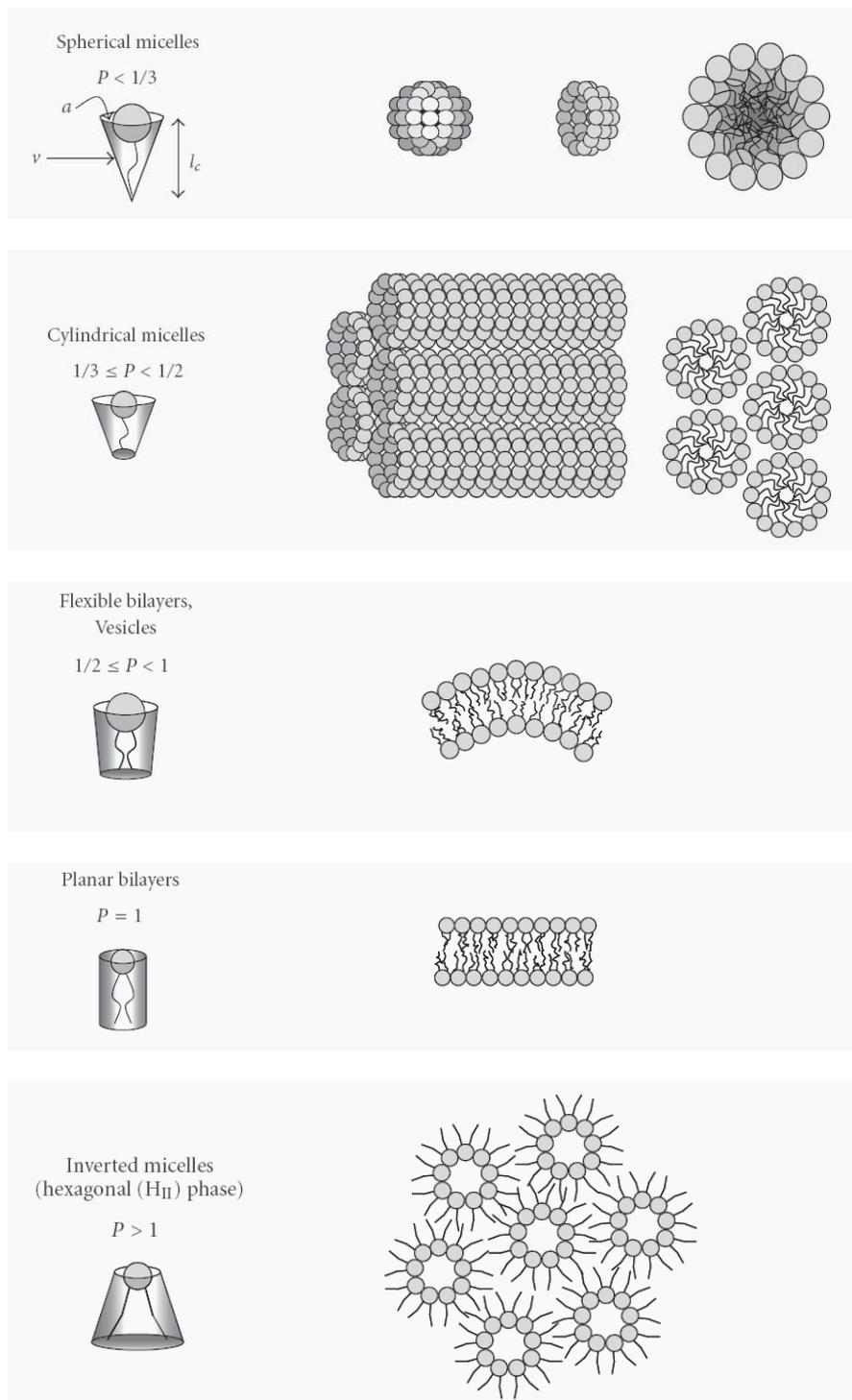
**equation 1.1**

$$P = \frac{v}{a \cdot l_c}$$

where  $v$ ,  $a$ , and  $l_c$  are volume, head group area and length of the amphiphile, respectively.

For surfactants with a broad head group and a short tail,  $P < 1/3$  and spherical micelles are formed, where the water forms the solvent phase. Shifting the packing parameters to higher values (by decreasing  $a_h$  and/or increasing  $l$ ) the surfactant microstructure changes from cylindrical aggregates [3, 4] ( $1/3 < P < 1/2$ ) to bilayers (lamellar phase [5, 6] and vesicles [5, 7-10] ( $P = 1$ ) and finally to the corresponding inverse structures as displayed in chart 1.1.

In the past the static behavior of these morphological structures has been studied intensively due to their central importance for technical and biological applications, which will be discussed in detail later. However, the dynamic properties of amphiphilic systems are only poorly understood and this applies also to the criteria determining the kinetics of structural reorganization processes of such systems.



**chart 1.1** Morphological structures predicted by the packing parameter [11].

Hence, the present work dealt with kinetical aspects in surfactant systems and the influence of parameters, such as charge, steric interactions, and temperature thereof. Three kinetic approaches of fundamental importance for technical applications will be investigated:

The kinetics of the **formation** of amphiphilic structures.

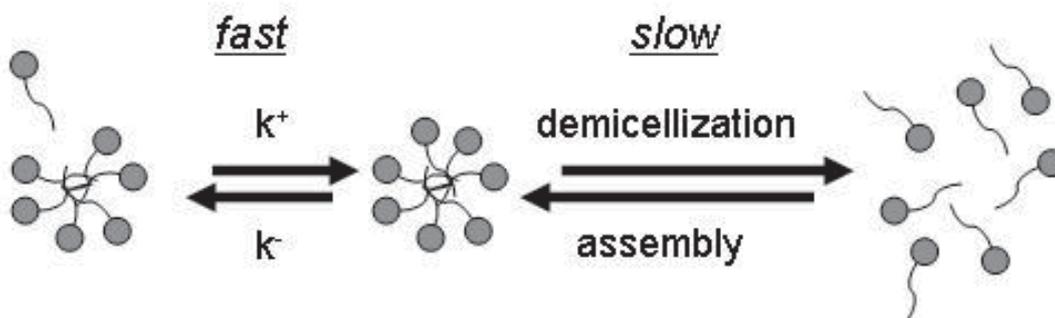
The kinetics of the **exchange** of solubilisate between surfactant aggregates.

On the one hand the morphological transition from micelles/microemulsions to metastable multilamellar vesicles and the formation of thermodynamically stable microemulsions was studied (where the latter is not a process of formation in the common sense because the amphiphilic structure of the starting and the final solution are MEs in both cases; it is therefore more appropriate to speak of equilibration of ME aggregates). The former one is of special technological interest since it has been known for some time now that metastable amphiphilic structures are controlled by the dynamics of their formation [12]. Studying the dynamics of morphological transitions in surfactant systems has become an increasingly active field of colloid research due to the improved experimental time resolution which offers the possibility to gain a more detailed picture of the structural changes involved [13]. Therefore the dynamics of these two formation processes were investigated i.e. by means of time-resolved SAXS measurements. Subsequently specific control of the metastable intermediate structures can be of key importance e.g. in template synthesis and formulations etc.

On the other hand the kinetic of the exchange of oil between ME aggregates was studied in detail. As amphiphiles two technical-grade surfactants were employed. Their structure and properties will be introduced in the following paragraph, where a brief introduction to microemulsions (definition, relevance and application) will be given. The last paragraph of the introduction will deal with the principles of vesicle formation and their importance for technical and biological tasks.

## Microemulsions

Microemulsions are thermodynamically stable dispersions of either water-in-oil (W/O) or oil-in-water (O/W), the stability being the result of the presence of a suitable surfactant [14, 15]. They have been studied intensively due to their enormous potential in many technical applications, e.g. in detergency, ternary oil recovery, formulation of cosmetic, agricultural or pharmaceutical products, and lately the preparation of nanoparticles [16-21]. ME aggregates saturated with hydrophilic (water) or hydrophobic (alkanes) solubilise, consist of small, spherical droplets, exhibiting radii in the range 3 - 30 nm [22]. MEs are thermodynamically stable (they form spontaneously and droplet size is independent of variations in preparation method [23]) which renders them most suitable as reaction medium for enzyme-catalyzed reactions [24, 25], the controlled and selective synthesis [26] and lately the preparation of nanosized particles [19-21].



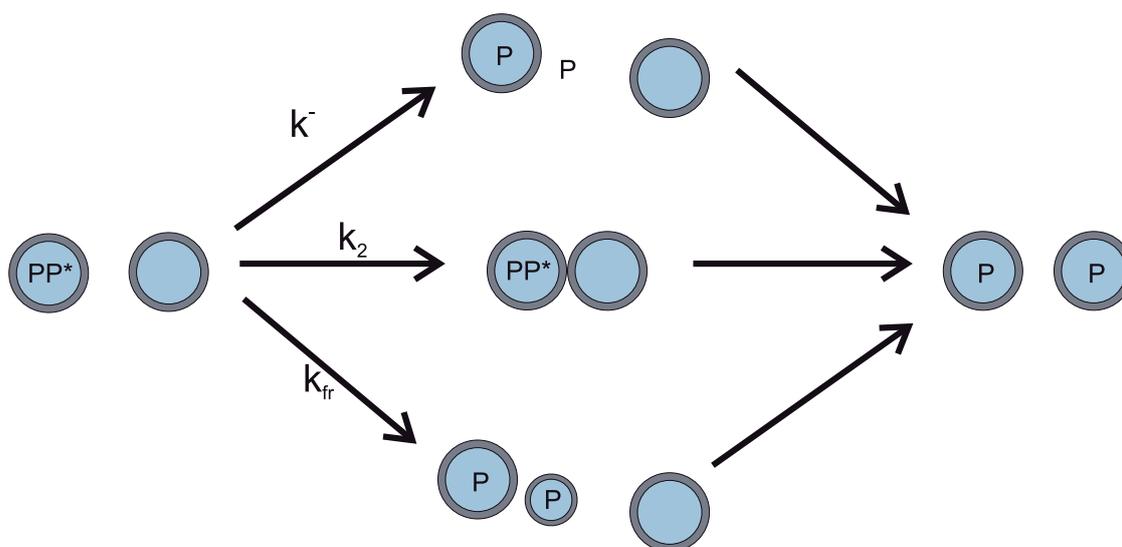
**chart 1.2** Surfactant exchange mechanism and micelle formation and break-down according to Aniansson and Wall [27] where  $k^+$  and  $k^-$  are the exit and entry rates of surfactant molecules (displayed as hydrophilic head group in blue and hydrophobic tail)

Quite a number of publications about micelle dynamics come from chemical relaxation experiments [28-31] that are a powerful means of determining the rates of various relaxation processes. Two well-separated relaxation times have been identified: a rapid relaxation  $\tau_1$ , which requires microseconds and a slower process  $\tau_2$  that occurs on a timescale of seconds or longer. A cartoon describing these two processes is presented chart 1.2. The fast process has been assigned to an association-dissociation process (exchange of individual surfactant molecules between micellar aggregates and the

continuous phase) [27, 32] and values for  $k^+$  and  $k^-$  have been determined, e.g. by performing T-jump experiments for a number of surfactants [33]. The slow process was attributed to the reestablishment of equilibrium through cooperative condensation or dissolution of surfactant monomers (creation and breakdown of entire micelles). However it is difficult, with data from relaxation experiments alone, to establish the mechanism associated with each rate particularly the one of the slow process [34].

When hydrophobic compounds are solubilised into the micellar core, another type of dynamics becomes important: the exchange of solute molecules between micelles/MEs [35]. In contrary to micelles, MEs contain a dispersed state consisting of domain of nanometer dimension. In addition the interfacial tension in a solution of MEs is very small [35]. Both structures represent a thermodynamically stable state of the respective system. Regarding solute exchange between the aggregates three different mechanisms have been proposed:

- \* The first one involves the exit of individual solute or surfactant molecules into the aqueous phase and the entry into another droplet. This exit-reentry mechanism (represented by the relaxation rate  $k^-$ ) becomes dominant when the water solubility of the species is sufficiently high.
- \* During the second mechanism the exchange of solute proceeds via collision of two micelles, followed by their separation. This collision-exchange-separation process, often called fusion-fission mechanism (and represented by the relaxation rate  $k_2$ ), has been suggested as the dominant exchange mechanism in nonionic MEs and nonionic surfactant micelles at high concentration or at elevated temperature [35].
- \* The third possibility for solute exchange involves the fragmentation of a micelle/ME into two sub-aggregates and the subsequent growth into normal micelles/MEs. This fragmentation-growth (represented by the relaxation rate  $k_{fr}$ ) has been suggested as a mechanism to explain rapid exchange between ionic micelles and MEs at high ionic strength [1].



**chart 1.3** Three different mechanisms for solute exchange between microemulsions (black circles refer to the surfactant monolayer; the hydrophobic solute is illustrated as gray-colored interior) where P is the fluorescent probe,  $k^-$  the exit rate for the probe molecule,  $k_2$  the exchange rate for the fusion-fission mechanism and  $k_{fr}$  the exchange rate for the fragmentation-growth mechanism [35]

These three different solute exchange mechanisms are displayed in chart 1.3. The circles refer to the ME (surfactant monolayer: black, solute: gray) and the letter P's refer to both a generic solute and a pyrene derivative that will give excimer fluorescence (see chapter 2.1.2) from aggregates containing two P molecules. The reaction rates  $k^-$ ,  $k_2$  and  $k_{fr}$  correspond to the exchange processes of the exit-reentry, the fusion-fission and the fragmentation-growth mechanism, respectively. In reaction kinetics, in general, the overall rate of a reaction is controlled by the rate of the slowest step. Depending on the number of participating molecules/aggregates etc. the order of reaction can be determined and from that an eligible fit function is selected. For the exit-reentry and the fragmentation-growth mechanism, only one aggregate is involved in the rate determining step. Therefore first-order kinetics is observed as shown in

**equation 1.2**

$$y = y_0 + A \cdot \exp(-k_{obs} \cdot t)$$

where  $y$  is the measured variable as function of the time  $t$ , for instance the decreasing excimer intensity,  $y_0$  the offset,  $A$  the amplitude of the signal and  $k_{obs}$ , the observed

relaxation rate. In many cases a mono-exponential fit function can be employed. The middle path of chart 1.3 will exhibit kinetics second order in ME concentration and can universally be expressed as

**equation 1.3**

$$y = y_0 + \frac{A}{1 + k_{obs} \cdot t}.$$

In the last 30 years only a limited number of authors presented results to understand the mechanism of the slow relaxation process described in chart 1.2. Hence there is a need to attempt to establish parameters of this process in more quantitative terms. As part of such an attempt, we investigate here O/W-microemulsions applying the stopped-flow technique [36, 37] which is a versatile method to study fast kinetics after a rapid mixing process with a time resolution of down to 1 ms, and has been employed in various studies before [38-44]. Details about the mode of operation are given in the experimental section 2.1.1.

Properties of microemulsion systems are to a large degree determined by the elastic properties of the amphiphilic monolayer [45, 46]. Description of MEs in that context is very simple, “*since, because of their low interfacial tensions, the leading term in the free energy should be given by the bending energy, which can be described in terms of two elastic constants, the mean bending modulus  $\kappa$ , and the Gaussian modulus  $\bar{\kappa}$* ” [47]. Here the influence of the composition, especially the cosurfactant concentration, was found to be of particular interest. In the course of this discussion the interrelation between the observed rate constants of the stopped-flow experiments and the macroscopic interfacial tension will be elucidated.

### ***Multilamellar vesicles***

Vesicles are closed amphiphilic bilayers where parts of the solvent is entrapped inside the aggregate and which are often formed in the low-concentration range. In general, vesicles can structurally be subdivided into two main classes. As a first case, they may just be comprised of one single bilayer, as displayed in the chart below and are then referred to as unilamellar vesicles/ ULVs with sizes between 4 – 20 nm for small unilamellar vesicles /SUV and 50 nm - 10  $\mu$ m for large unilamellar vesicles/ LUVs (denotation used in this work see chart 1.4). In addition to the first case there exists also the possibility of multilamellar vesicles/ MLVs (see chart below) where one has various concentric shells of vesicles, thus, these phases are also sometimes called ‘onion phases’ [48]. Vesicles especially from phospholipids (ULVs in this case are often called liposomes) have been investigated widely [10, 49]. They serve as model(s) for membranes and allow therefore studying the basic mechanisms of membrane function. Due to their closed interface they are convenient architectures for encapsulation necessary for pharmaceutical, cosmetic and chemical application.

The kinetic of morphological transitions in amphiphilic systems is important for the understanding of many applications but so far no comprehensive study has been done. The formation of vesicles is not a thermodynamically controlled process. In many cases the final vesicle phase is not in thermodynamic equilibrium but instead long-lived metastable structures are observed. Accordingly the properties of such systems depend strongly in the preparation of the sample and are therefore controlled by the kinetics of their formation process. The knowledge of the dynamics during the formation of the vesicles is crucial to influence the properties of the final state which is especially important in industrial applications.

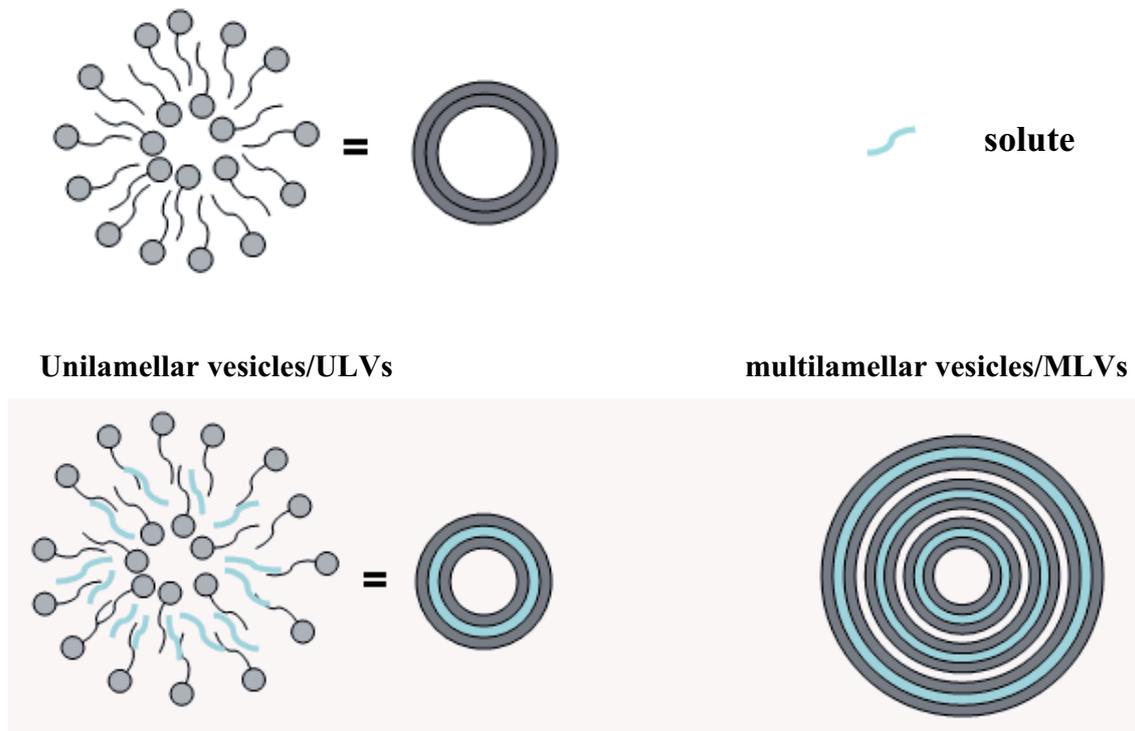
As already mentioned above, the elastic properties of amphiphilic monolayers and bilayers, as they are typically formed in self-assembling structures, are an important factor in understanding their properties [47] and can be described by a spontaneous curvature  $c_0$  and two elastic constants  $\kappa$  and  $\bar{\kappa}$  associated with the mean curvature and the Gaussian curvature, respectively. These parameters are very important in the determination of the structure and phase sequences in surfactant systems, i.e. droplets or sponge-like structures and are interrelated in the harmonic approximation of the

continuum bending free energy,  $F_b$ , of a monolayer or bilayer established by Helfrich in 1973 [50]:

**equation 1.4**

$$F_b = \int dA \left[ \left( \frac{\kappa}{2} \right) \cdot (c_1 + c_2 - 2c_0)^2 + \bar{\kappa} \cdot c_1 \cdot c_2 \right]$$

where  $c_1$  and  $c_2$  are the principal curvatures of the amphiphilic film,  $\kappa$  and  $\bar{\kappa}$  are the mean and Gaussian bending moduli and  $A$  is the surface area. With this approximation one can calculate the energy of the different topologies of a surfactant film and thus determine the most stable structures. The expression for  $F_b$  in equation 1.4 is essentially an expansion of the free energy for small curvatures and is correct in the limit of radii of curvatures that are large compared with the thickness of the film [51].



**chart 1.4** Illustration of unilamellar and multilamellar vesicles