**Title**

# Vesicles and lamella: outcome of the changing formation path of a sodium N-lauroylsarcosinate hydrate/1-decanol/water system

**Authors:**

1. Nasima Akter (Corresponding Author)

School of Applied Physics, Faculty of Science and Technology,

Universiti Kebangsaan Malaysia, 43600 UKM, Bangi, Selangor, Malaysia.

Tel: +60389214256; Fax: +60389269470

Email: nasima.physics@yahoo.com; nasima.physics@gmail.com

2. Shahidan Radiman

School of Applied Physics, Faculty of Science and Technology,

Universiti Kebangsaan Malaysia, 43600 UKM, Bangi, Selangor, Malaysia.

Tel: +60389213383; Fax: +60389269470; Email: shahidan@ukm.my

3. Faizal Mohamed

School of Applied Physics, Faculty of Science and Technology,

Universiti Kebangsaan Malaysia, 43600 UKM, Bangi, Selangor, Malaysia.

Tel: +60389215475; Fax: +60389269470; Email: faizalm@ukm.my

4. Mohammad Imam Hasan Reza

Molecular Biology Lab, Department of Botany, University of Chittagong, Chittagong 4331, Bangladesh. Tel: +88031726311; Fax: +88031726310; Email: rezamih@gmail.com

**Vesicles are closed bilayer that enclosed a part of the continuous phase inside the core. In spherical shape, it attains the minimum free energy state. Conversely, lamella with maximum free energy remains in planer bilayer shape in the colloidal dispersion. Even with the same amphiphile concentration the colloidal structures depends on different parameters, many of these already addressed in different reports. However, the effect of mixing procedure as a formation path is unidentified. Here we reported water in 1-decanol and 1-decanol in water; these two different mixing procedures yield vesicles and lamella at the same point of the phase diagram of a sodium N-lauroylsarcosinate hydrate/1-decanol/water system. It was found that the favorable and unfavorable contact of water with the weak tertiary ammonium cation in amino-acid head-group plays the crucial role in this process. Moreover, this weak cationic property of this amphoteric surfactant can be exploited to carry DNA for gene therapy with a nontoxic system instead of cationic.**

[Keywords: Mixing procedure, lamella, vesicle, aggregate structure, phase diagram]

There have been relentless efforts to understand the colloidal structures and their potentials in biological sciences since the beginning of their discovery[1-4]. The principal aims of such attempts are to improve the drug efficacy, membrane property study, gene therapy, and cosmetic formulation. Amphiphile molecules having a polar head and a nonpolar tail have an affinity to form different aggregate structures when the surrounding environment changes. The colloidal system exhibits competition between different processes, giving rise to a special pattern formation. Among different environmental parameters, sonication, pH[5], salt[6-8], temperature effects[9-11] are already well understood. However, the effect of the mixing procedure in the colloidal structures has remained a curiosity. In this context, it is important to elucidate its effect in colloidal structure.

In this study, we used one single chain amino acid-based surfactant; sodium N-lauroylsarcosinate hydrate (SNLS), 1-decanol and water. Due to a bulky ionic polar head and strong electrostatic repulsion, the single chain ionic surfactant always forms micelles in pure water[12]. Furthermore, like other fatty acid vesicles, it forms bilayers or vesicles in the presence of other neutral amphiphiles, which doubles the hydrocarbon volume[13-15]. An ion pair formed by the association of two headgroups of ionic and neutral amphiphiles induces a bilayer structure. A stable vesicle lobe was found in this system, and the detail phase behavior of this system discussed elsewhere[16].

An attempt has been made to understand the effect of mixing procedure on the formation of aggregate structure. Previously, Marques[17] studied the vesicle formation path of one catanionic system, which involved mixing of solutions, mixing of solids and dilution of concentrated samples. Dan and coresearchers[18] investigated the effect of mixing on the morphology of cylindrical micelles. They reported the increasing spontaneous curvature of an amphiphile leading to a first-order morphology transition from threadlike micelles to a branched network. However, we studied the vesicle formation path, involving two mixing

procedure: (a) 1-decanol in water and (b) water in 1-decanol at the same point of the phase diagram. Atypical behavior was observed: where two different aggregate structures yield in two different mixing procedures.

Generally, a mixture of water-soluble and water-insoluble surfactant results in vesicle formation after immersion in water[19]. With SNLS and 1-decanol, vesicles were formed, when the mixture of the two amphiphiles (at a particular molar ratio) is immersed into water. On the other hand, lamella has formed when adding 1-decanol in the micelle solution. It was reported that 1-decanol induces a lamellar structure in some systems[20,21]. In fact, variation of adsorbed water on the monolayers determines the optimal head group area. In first case, 1-decanol inserts tearing the network of strongly adsorbed water around micelle. Generally, insertion of long chain alcohol reduces the headgroup area, which induced formation of lamella. However, in the case of the second mixing procedure, the monolayers forms in the presence of water, therefore, water adsorbed homogeneously on the headgroup area.

Among all bilayer structures, vesicles achieved considerable importance owing to their cell mimic properties and widespread applicability in practical and theoretical field. Until now, many works have been devoted to improve the understanding of this system. In this present study, our aim was to investigate whether changing the formation path can induce the aggregate structures. In fact, as we have shown from results, the changing environment plays a important rule on aggregate structures. Therefore, it can be believed that this fundamental result will contribute in part to colloid and interface science.

## RESULTS

In order to obtain a clear understanding of the dynamics of vesicle formation, we prepared our samples following two different mixing protocols at the same point (shown in Table1 in 92 wt% water) of the phase diagram (the single phase region III)[16], they are as follows:

1. 1-decanol in water, i.e., mixing the surfactant in water and then adding 1-decanol, and

2. Water in 1-decanol, i.e., surfactant were mixed with 1-decanol first and then adding water.

**Table-1** Formation of vesicle and lamella in different molar ratios of the amphiphiles

| Amphiphile concentration ($mmol/dm^3$) | Molar ratio of amphiphiles | | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 1:2.10 | | 1:2.15 | | 1:2.32 | | 1:2.47 | | 1:2.65 | | 1:2.90 | | 1:3.10 | | 1:3.40 | | 1:3.39 | |
| 0.43 | V | V | V | V | L | V | L | V | L | V | L | V | L | V | L | V | L | V |

We noticed that above the molar ratio 1:2.15, the macroscopic appearance of the first sample was nearly opaque and less fluid, and that of second sample was bluish with increasing fluidity. Moreover, the first sample shows a rainbow texture between cross polarizers, while the other sample shows floating birefringent. These results are designated as L (for lamella) and V (for vesicle) in table-1. These results also verified using small angle scattering X-ray experiment (Fig. 1).

In case of protocol 1, the corresponding spectra of the scattered intensity versus scattering vector q showing four Bragg reflections with the d-spacing ratios 1:1/2:1/3:1/4 (Fig. 1a).

The first Bragg peak is higher than the second one, which is the characteristic of lamellar periodicity in the case of fully hydrated sample in $L_\alpha$ phase[22,23]. It is also notable, that the first Bragg peak appears at very low q: in other words the d spacing is very high, which is due to low concentration of the surfactant, where we used only 8wt% amphiphiles (3.5% SNLS + 5.5wt%1-decanol) surfactant. It is recognized that d-spacing increases with the increasing water[22,24-25]. From the small angle X-ray scattering investigation of the vesicle dispersion (SNLS/1-decanol molar ratio = 1:2.32 in 92 wt% water, protocol 2), it was found that the

scattering spectra consists of two peaks, a broad scattering peak and a small sharp peak (Fig. 1b). The broad peak represents a single bilayer[26,27] and the second sharp peak arises from the interparticle interference[28].

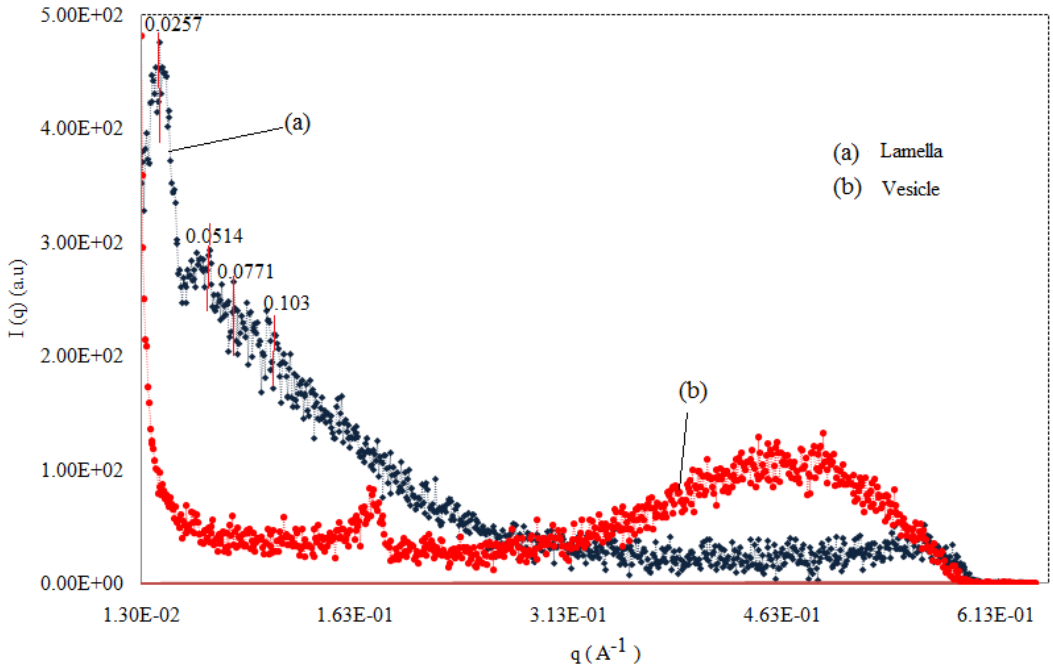


**Figure 1.** SAXS spectra of a sample with molar ratio 1:2.32 system in 92 wt% water, where (a) represents the scattering peak for lamella where, the vertical straight line indicates the lamellar repeated distances (using 3D view indexing) and, (b) represents the scattering peak of vesicle solution. The results are shown after background subtraction.

In order to reveal the aggregate structures, which are beyond the scope of a small angle scattering x-ray experiment, we characterized the samples using polarized microscope and TEM. These results have shown in Fig.2, where cloudy pattern observed under polarized microscope (shown in Fig. 2a), and the corresponding rainbow texture of the sample through cross polarizing filters (shown in Fig. 2b) are the proof of the lamellar $L_\alpha$ phase. When observed through cross polarizing filters, floating birefringent was observed in case of vesicular sample (Fig. 2c). The TEM image of the lamellar phase was dark. This darkness is attributed to the parallel orientation of the lamellar stakes on the surface of the grid, which

prevents the electron beam to pass through the aggregate structures. On the other hand, the internal aqueous core and bright boundary is visible in the vesicle image (Fig. 2d). The microscope image and visual observation through cross polarizers are direct evidence of this dissimilarity.

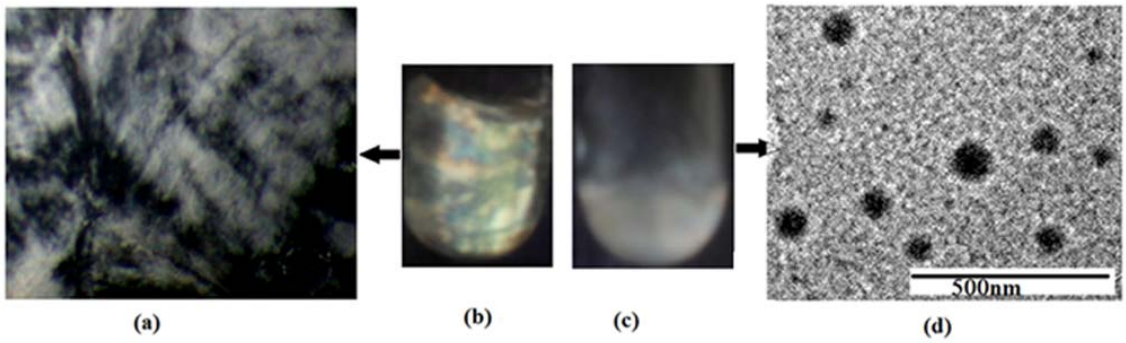


**Figure. 2.** (a) optical microscope image of a lamellar ($L_\alpha$) phase, (b) the corresponding image of the sample in a lamellar phase through cross polarizing filters, (c) the sample in vesicle phase through cross polarizing filters and, (d) transmission electron micrograph of the vesicles at the same point of the phase diagram (total amphiphile concentration 0.43m mol/$dm^3$ and molar ratio is 1:2.32).

To explore the reason of such variation we investigated the FTIR spectra. Fig.3(a), 3(b), 3(c) and 3(d) are the FTIR spectra of sodium N-lauroylsarcosinate hydrate, 1-decanol, lamella and vesicle. In case of lamella and vesicle, maxima were found at 1656 $cm^{-1}$ (amide region); however, in case of lamella, the peak was observed with falling intensity. These peaks are shifted from a split peak in the surfactant at 1640 and 1648 $cm^{-1}$ (Fig. 3a). The shift is attributed to the hydrogen bonding between the C=O of the amide group and the alcoholic hydroxyl group. Tenaciously bound water also contributes to this bonding. The reduction of

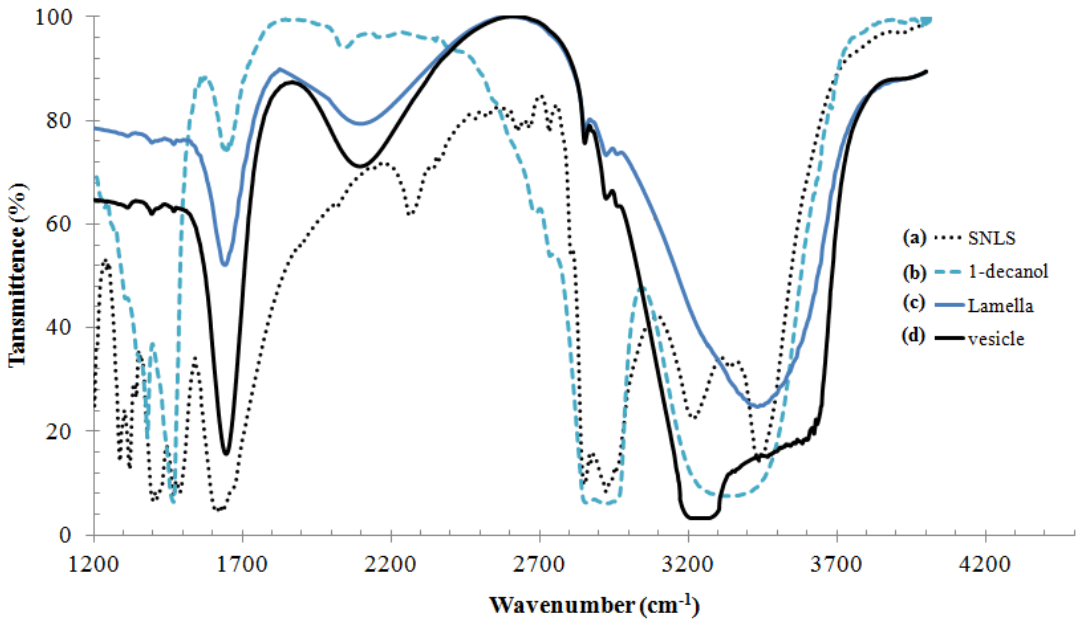


**Figure 3.** FTIR spectra of (a) sodium N-lauroylsarcosinate hydrate, (b) 1-decanol, (c) lamella and (d) vesicles in solution for the molar ratio of 1:2.32 systems in 92 wt% water.

bound water onto the monolayer weakens the peak strength in case of lamella. Strong hydrogen bonding yields a significant band broadening in region of 3400 $cm^{-1}$. Both OH stretching vibrations from the alcohol group and adsorbed water contributes to this broadening. However, a hydrogen band at 3304-3229 $cm^{-1}$ is an evidence of the tertiary ammonium cation ($R_3N^+H$). Owing to the amino acid group, this anionic surfactant often behaves as slightly cation-active[29]. In the case of the lamellar solution, a comparatively narrow peak was observed at 3400 $cm^{-1}$, while peak at 3304-3229 $cm^{-1}$ was overlapped with a hydroxyl group of 1-decanol. The diminution of peak shape is due to the significant reduction of adsorbed water from the hydrophilic surface (Fig. 4a).

In the vesicle solution, before immersion in water strong hydrogen bonding was occurred between ionic and neutral head groups. Addition of water hydrates the hydrophilic part of the monolayer, which reflects from the broad hydration peak. The strong asymmetric and symmetric stretch of methyl groups of SNLS and 1- decanol appears as very weak stretching vibration at the peak value 2956 $cm^{-1}$ and 2856 $cm^{-1}$ in both cases, these changes are ascribed to the association of more acyl chains, which weaken the peak strength (Fig. 4b).

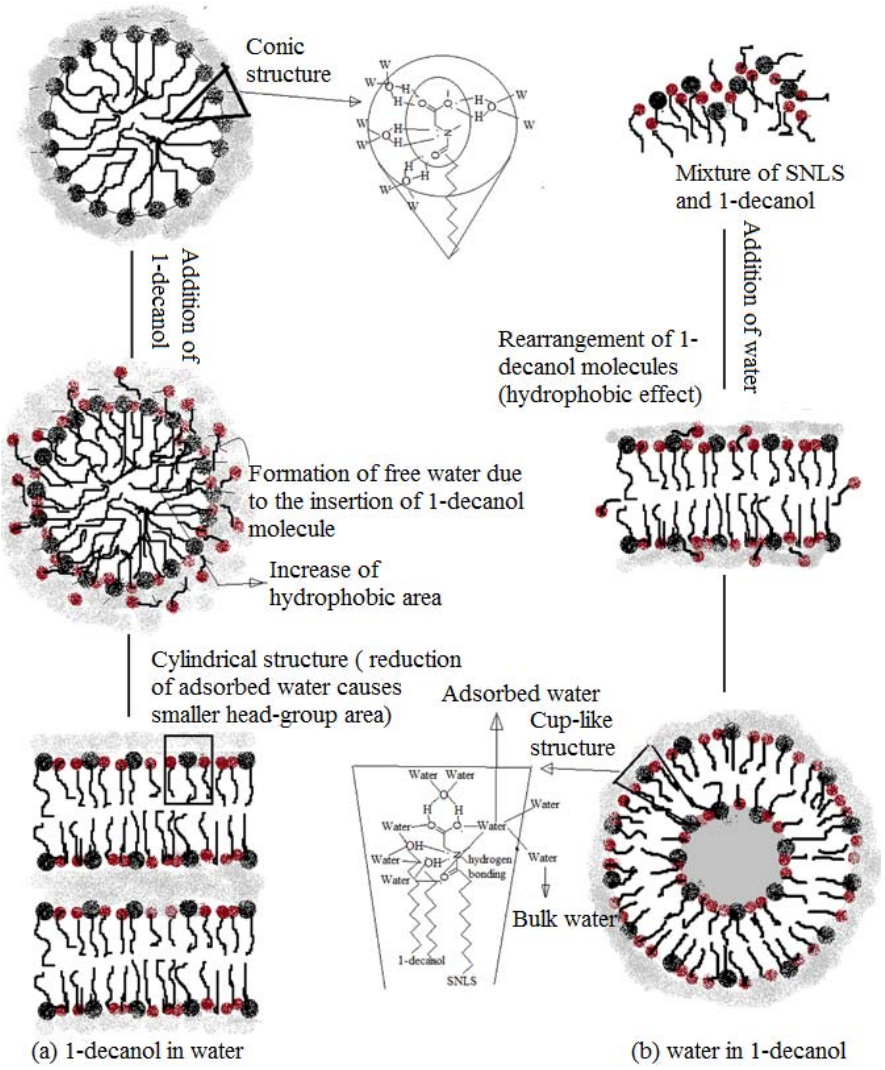


**Figure 4.** Schematic representation of the evaluation of the mixed amphiphiles of the two protocols. (a) the formation of lamella (b) the formation of vesicles, and in the middle the conic and cylindrical structures are shown with their effective geometrical area.

## DISCUSSION

The attractive dispersion force between hydrocarbon chains and the repulsive forces between headgroups determine the aggregate structure in colloidal dispersion. In a charged system, this repulsive force arises from charge- charge repulsion and head group hydration. A useful model to predict the colloidal aggregate structure is "molecular packing parameter" introduced by Israelachvili[30]. The packing parameter is defined as the ratio of the volume of the hydrocarbon tail of the surfactant in the core ($v_c$) and the product of the optimal head group area ($a_0$) and the critical chain length of the tail ($l_c$).

$$P = v_c / a_0 l_c$$

The spherical micelles are formed at $0<P<1/3$ and the cylindrical micelles are formed at $1/3<P<1/2$, for $½<P<1$, vesicles are formed, at $P\approx1$ formed lamella and for $P>1$ reverse micelles are formed.

Our results indicate that morphologically two different types of aggregate structure achieved for two different mixing procedures. The underlying reason might be, in the first mixing procedure, the relative amount of bound water started to decrease due to the addition of 1-decanol in the micelle solution, which further reduces the head group hydration. It is well known that the formation of free water is inversely proportional to chain length and concentration of amphiphiles[31]. Being a long chain molecule, 1-decanol at increasing concentration, freed more bound water molecules from micelle surface. With a very small headgroup area ($a_0 < 20\ A^0$) and strong hydrogen bonding property, 1-decanol molecules approach closer to the ionic headgroup[32]. Hence, the more the 1-decanol molecules insert, the smaller the headgroup area become. From the FTIR spectra, the tertiary ammonium peak appears very weak and the mean peak shifted to lower frequencies due to the substitution of water molecules, and hydrogen bonding between surfactant headgroup and 1-decanol molecules. The acyl chains of alcohol molecules penetrate into the palisade layer, arranged

with hydrophobic group towards the micelle core, and the hydrophilic parts strongly attached to the head groups and formed mixed micelle[33]. Generally, insertion of long chain alcohol reduces the head group area[34]. Despite for each $–CH_2$- and $–CH_3$ a gain of ~630 cal/ mol and 2100 cal/mol respectively, is achieved, when the molecule (1-decanol) transferred from water to non polar micelle core[35,36], such a gain also was not enough to favor vesicle formation in first protocol, when the molar ratio of the amphiphile mixture was 1:2.32 to 1:3.40. The larger hydrophobic area and very compact hydrophilic area makes the area cylindrical in shape (i.e., the packing parameter value $P \approx 1$), the cylindrical structure is responsible for the formation of lamella. From a similar study, it is reported that addition of 1-decanol decreases the radius of curvature in one sodium dodecyl sulphate (SDS)/water system[37].

In the case of second procedure, in the SNLS/1-decanol mixture, the 1-decanol molecules prearranged themselves within the ionic amphiphiles. The immersion of the mixture in water hydrated the polar group and increased the relative head group area. Comparing the FTIR results of sodium N-lauroylsarcosinate hydrate and vesicle dispersion, it is noted that the tertiary ammonium cation is strongly hydrated in vesicle solution, where broad vesicular peak 3236-3300 $cm^{-1}$ is shifted from the 3236 $cm^{-1}$ narrow peak of surfactant. Worth mentioning that, this mild cationic part plays the principle rule in this deviation. The acyl chains become stiffer because of the presence of water molecules in its immediate vicinity, and hiding themselves from strongly adsorbed water as well as the hydrophilic surface, expand due to the hydration force. Both repulsive force and hydration force in this case is not as strong as micelle due to the presence of excess neutral amphiphiles. The simultaneous expansion and contraction both cannot transfer the system conic or cylindrical structure, owing to the intermediate adherence to the bound water. The system turns into a cuplike structure, which is the basis of vesicle formation.

As our main purpose is to prepare a suitable drug delivery system, and special interest in vesicular aggregate structure, we emphasize about the knowledge of mechanism of vesicle formation. Therefore, we studied the path of vesicle formation. Depending on two different mixing protocols as formation paths, strikingly different results were achieved. Generally, lamella formed owing to an energetically unfavorable contact with the surrounding water pool, giving rise to a lateral tension. This is in contrast to vesicular system, which is essentially a tension-free system, where the higher adsorbed water giving rises to a higher curvature, which triggers the colloidal system into vesicular structure. Despite the similar packing of the acyl chains, difference in bound water with tertiary ammonium cation is responsible for such occurrence. Hence, it can be suggested that vesicle formation depends not only on the appropriate choice of amphiphiles, solubility property, temperature and molar ratio but also on the correct mixing procedures of the amphiphiles.

Our results constitute essential information that the hydrogen bonding property of the tertiary ammonium cation has a great influence on the surrounding environment. This important property can be exploited by the incorporation of important macromolecules; such as DNA for gene therapy. This will essentially be precipitation free, since no electrostatic attraction will be present in this system. Moreover, incorporation of this vesicle in food colloids; such as kappa-carrageenan gel network will facilitate for oral drug delivery. In both case a hydrogen bond will results in between the hydrogen atom attached to the carbon and a lone pair electron of nitrogen. It was reported that amphoterics with repeated amino and carboxylic group show binding of polyvalent cations, such as NTA and EDTA[38] and also in another report is was shown that amino acid based amphoteric molecules can be used for siRNA delivery[39]. Considering such potential advantages, morphological study of this system requires increasing attention. Furthermore, it can be assume that, in future this fundamental

result will play an important role for deciding the appropriate mixing procedure as a path of formation of colloidal dispersions.

## METHODS

Sodium N-lauroylsarcosinate hydrate ($C_{15}H_{28}NNaO_3$ x $H_2O$) was purchased from TCI (Japan), 1-decanol with 97% purity was purchased from Fluka-Chemica (Switzerland). Deionized water was used to prepare all the samples. To obtain mixed assemblies i) required amount of surfactant (SNLS) was mixed with water and then 1-decanol was added with the mixture; and ii) same amount of amphiphiles were measured in the glass tube and then required amount of water was added. In both cases, the concentration and molar ratio of amphiphiles remain same. The resulting suspension was sonicated for 10 minutes at 20 ºC using a bath type sonicator. The homogenous dispersions were then centrifuged at 4,000 rpm for 10 min to see the phase separation. Before analysis, all samples were left undisturbed for equilibration for several days.

A polarizing light microscope from Meiji Techno Co. Ltd., Japan, equipped with a camera (Nikon, Model–E995) was used to observe the anisotropic phases of the sample. Samples for light microscope were prepared by placing a droplet on a microscope slide and gently pressing a cover glass on top of the droplet. Photographic images of the textures were recorded with the camera.

The characterization procedure of transmission electron microscope, small angle x-ray scattering, and Fourier transform infrared spectra is discussed elsewhere[16].

**Reference**


1. Aranda-Espinoza, H. *et al.* Electrostatic repulsion of positively charged vesicles and negatively charged objects. *Science* **285,** 394-397 (1999).
2. Laughlin, R.G. Surfactant phase science. *Curr. Opin. Colloid Interface Sci.* **1,** 384-390 (1996).

3. Hostetler, M.J. & Murray, R.W. Colloids and self-assembled monolayers. *Curr. Opin. Colloid Interface Sci.* **2,** 42-50 (1997).

4. Palberg, T. Colloidal crystallization dynamics. *Curr. Opin. Colloid Interface Sci.* **2,** 607-614 (1997).

5. Quemeneur, F., Rammal, A., Rinaudo, M. & Pe´pin-Donat, B. Large and giant vesicles 'Decorated' with chitosan : effects of pH, salt or glucose stress, and surface adhesion. *Biomacromolecules* **8,** 2512-2519 (2007).

6. Fujiwara, M., Grubbs, R.H. & Baldeschwieler. J.D. Characterization of pH-dependent poly(acrylic acid) complexation with phospholipid vesicles. *J. Colloid Interface Sci.* **185,** 210-216 (1997).

7. Schurtenberger, P., Mazer, N. & Kaenzig, W. Micelle to vesicle transition in aqueous solutions of bile salt and lecithin. *J. Phys. Chem.* **89,** 1042-1049 (1985).

8. Walter, A., Kuehl, G., Barnes, K. & Waerdt, G.V. The vesicle-to-micelle transition of phosphatidylcholine vesicles by nonionic detergents: effect of sodium chloride, sucrose and urea. *Biochim. Biophys. Acta (BBA)- Biomembranes* **1508,** 20-33 (2000).

9. Tschierske, C. Amphotropic liquid crystals. *Curr. Opin. Colloid Interface Sci.* **7,** 355-370 (2002).

10. Yin, H., Lin, Y., Huang, J. & Ye, J. Temperature-induced vesicle aggregation in catanionic surfactant systems: the effects of the headgroup and counterion. *Langmuir* **23,** 4225-4230 (2007).

11. Guo, C., Zhang, R., Jiang, L. & Liu, T. Invesigation on the temperature effect of a mixed vesicle composed of polydiacetylene and BODIPY 558. *Colloids Surf. B* **60,** 41-45 (2007).

12. Zemb, T., Dubois, M., Deme, B. & Gulik-Krzywicki, T. Self-assembly of flat nanodiscs in salt-free catanionic surfactant solutions. *Science* **283,** 816-819 (1999).

13. Gradzielski, M., Muller, M., Bergmeier, M., Hoffmann, H. & Hoinkis, E. Structural and macroscopic characterization of a gel phase of densely packed monodisperse, unilamellar vesicles. *J. Phys. Chem. B* **103,** 1416-1424 (1999).

14. Morigaki, K. & Walde, P. Fatty acid vesicles. *Curr. Opin. Colloid Interface Sci.* **12,** 75-80 (2007).

15. Vlachy, N. *et al.* Spontaneous formation of bilayers and vesicles in mixtures of single-chain alkyl carboxylates: effects of pH and aging and cytotoxicity studies. *Langmuir* **24,** 9983-9988 (2008).

16. Akter, N., Radiman, S., Mohamed, F., Rahman, I.A. & Reza, M.I.H. Ternary phase behaviour and vesicle formation of a sodium N-lauroylsarcosinate hydrate/1-decanol/water system. *Sci. Rep.* 1, 71; DOI:10.1038/srep00071 (2011).

17. Marques, E.F. Size and stability of catanionic vesicles: effects of formation path, sonication and aging. *Langmuir* **16,** 4798-4807 (2000).

18. Dan, N., Shimoni, K., Pata, V., Danino, D. & Anomalous, Y. Effect on the morphology of cylindrical micelles. *Langmuir* **22,** 9860-9865 (2006).

19. Lasic, D.D., Kidric, J. & Zagorc, S. A simple method for the preparation of small unilameller vesicles. *Biochim. Biophys. Acta* **896,** 117-122 (1987).

20. Friman, R., Danielsson, I. & Stenius, P. Lamellar mesophase with high contents of water: X-ray investigations of the sodium octanote-decanol-water system. *J. Colloid Interface Sci.* **86,** 501-514 (1982).

21. Ekwall, P., Mandell, L. & Fontell, K. Ternary systems of potassium soap, alcohol, and water: I. phase equilibria and phase structure. *J. Colloid Interface Sci.* **31,** 508-529 (1969).

22. Worakitkanchanakul, W. *et al.* Aqueous-phase behavior and vesicle formation of natural glycolipid biosurfactants, mannosylerythritol lipid-B. *Colloids Surf. B: Biointerfaces* **65,** 106-112 (2008).

23. Pabst, G. Global properties of biomimetic membranes: perspectives on molecular features. *Biophys. Rev. Lett.* **1,** 57-84 (2006).

24. Zhang, G.D., Chen, X., Xie, Y.Z., Zhao, Y.R. & Qiu, H.Y. Lyotropic liquid crystalline phases in a ternary system of 1-hexadecyl-3-methylimidazolium chloride/1-decanol/water. *J. Colloid Interface Sci.* **315,** 601-606 (2007).

25. Li, C. *et al.* Self-assembly of lyotropic liquid crystal phases in ternary systems of 1,2-dimethyl-3-hexadecylimidazolium bromide/1-decanol/water. *J. Colloid Interface Sci.* **342,** 354-360 (2010).

26. Hauser, H. Naturally occurring amphiphiles: aspects of their phase behavior. In *Reverse Micelles* (eds. Luisi, P. L. & Straub, B. E.) 37-53 (Plenum Press, New York, 1984).

27. Riske, K.A., Amaral, L.Q., Dobereiner, H.G. & Lamy, M.T. Mesoscopic structure in the chain-melting regime of anionic phospholipid vesicles: DMPG. *Biophy. J.* **86,** 3722-3733 (2004).

28. Feigin, L.A. & Svergun, D.I. *Structure Analysis by Small-Angle X-Ray and Neutron Scattering* (ed. Taylor G. W.) (Plenum Press, New York, 1987).

29. Hummel, D.O., Baum, A., Liu, M., Pastura, A. & Weber, A. *Analysis of Surfactants: Atlas of FTIR-Spectra with Interpretations* (Hanser Gardner Publications, Germany, 1996).

30. Israelachvili, J.N. *Intermolecular and Surface Forces* (Chapter 16, Academic Press, London, 1992).

31. Nagarajan, R. Molecular packing parameter and surfactant self-assembly: the neglected role of the surfactant tail. *Langmuir* **18,** 31-38 (2002).

32. Israelachvili, J.N. The interdependence of intra-aggregate and inter-aggregate forces. *Journal de Physique* **46,** 2125-2136 (1985).

33. Ogino, K. & Takeshita, N. Electrophoresis and diffusion studies of the solubilisation of polar and nonpolar oily compounds with sodium dodecyl sulphate. II. *Bull. Chem. Soc. Jpn.* **53,** 611-615 (1980).

34. Greipernau, B. *et al.* 1-alkanols and membranes: a story of attraction. *Biochim. Biophys. Acta* **1768,** 2899-2913 (2007).

35. Tanford, C. *The Hydrophobic Effect* (Wiley, New York, 1980).

36. Lasic, D.D. *Liposomes: from Physics to Applications* (Elsevier: Amsterdam, the Netherlands, 1995).

37. Hendrikx, Y. & Charvolin, J. Structural phase transformations induced by the addition of decanol to the hexagonal phase of the binary sodium decyl sulphate/water system. *Liq. Cryst.* **11,** 677-698 (1992).

38. Fainerman, V.B., Möbius, D. & Miller, R. *Surfactants: Chemistry, Interfacial Properties, Applications.* (Elsevier, 2001).

39. Adami R.C. *et al.* An Amino Acid-based Amphoteric Liposomal Delivery System for Systemic Administration of siRNA. *Mol. Therapy* **19,** 1141–1151 (2011).



**Acknowledgements**

We thank Orowa A. Sikder (St. Catherine's College, University of Oxford) for valuable comments. We also like to thank Universiti Kebangsaan Malaysia for the financial support of this research work through the grant number STGL-007-2010/11 and the UKM Zamalah Fellowship to NA.

## Author Contributions

N.A. conceived the research, did the artwork. N.A. and S.R. drafted the manuscript. F.M. & M.I.H.R. contributed intellectually. All authors contributed to the analyses. All authors discussed the results and contributed to the revision of the final manuscript.

## Competing Financial Interests Statement

The authors declare no competing financial interests.