

Hydrodynamic Interactions in Charged Vesicles Suspensions

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Abstract We have measured the short-time dynamics and the structure of charged liposome dispersions upon increasing the volume fraction. Although the structural properties of the suspensions suggest an interparticle potential not purely repulsive, the hydrodynamic theory of Beenakker-Mazur can explain the experimental hydrodynamic functions of the dispersions. This result suggests the generality of the theory, which up to now, only had been tested in pure repulsive systems.

1 Introduction

The complete description of colloidal interactions includes both, direct and hydrodynamic interactions. Considerable progress has been made in the description of both interactions using hard-sphere colloids suspended in apolar, nearly index-matching fluids (Segré et al. 1995; Pusey et al. 1997). Indeed, to access information on both interactions requires the measurement of the static and dynamic structure factor, which are easily accessible by static and dynamic light scattering. However, these exper-

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imental methods suffer from the need of measuring single scattered light, which is difficult to achieve when the refractive index of the colloidal particles differs considerably from the one of the solvent. This, however, is the case for most charged colloidal systems, where the direct interactions are dominated by Coulomb-repulsion. Indeed, measurement on the hydrodynamics of charged particles mostly involved silica particles suspended in various organic solvent mixtures (Philipse and Vrij 1988; Härtl et al. 1999; Rojas et al. 2003; Phalakornkul et al. 1996; Riese et al. 2000) (none of them pure water), where the refractive index is nearly matched to the one of the solvent.

To gain an understanding of interplay between direct and hydrodynamic interactions in relatively dense charged colloidal systems we thus investigate the static and dynamic properties of aqueous suspensions of charged liposomes. These systems nearly fulfill conditions, index and density matching. Moreover, hydrodynamic theories have only been tested in pure repulsive systems. In fact, more than twenty years ago, Genz and Klein addressed the relevance of testing these theories in systems showing other kind of direct interactions, like attractive ones (Genz and Klein 1991). In this chapter, we will study the validity of the hydrodynamic theories in dispersions of charged liposomes, which show static structure factors that cannot be explained by a purely repulsive potential (Haro-Pérez et al. 2009).

2 Hydrodynamic Interactions

The dynamic structure factor depends on the direct interaction potential between the colloidal particles and on the indirect hydrodynamic interactions mediated by the solvent. Hydrodynamic interactions arise from the flow patterns generated in the surrounding fluid by the moving particles. The effect of direct and hydrodynamic interactions on the particle diffusion is described by the well-known expression $D_{eff}/D_0 = H(q)/S(q)$ (Pusey 1991), where $D_{eff}(q)$ and D_0 are the effective and the single particle diffusion coefficient, respectively. The hydrodynamic function, $H(q)$, contains the configuration-averaged effect of hydrodynamic interactions on the particle dynamics.

The hydrodynamic function $H(q)$ is connected to the mobility tensor $\tilde{\mu}_{ij}(\{\vec{r}^N\})$ by

$$H(q) = \frac{S(q)D_{eff}(q)}{D_0} = \frac{\kappa_B T}{ND_0} \sum_{i,j=1}^N \langle \hat{q} \cdot \tilde{\mu}_{ij}(\{\vec{r}^N\}) \cdot \hat{q} e^{i\hat{q} \cdot (\vec{r}_i - \vec{r}_j)} \rangle \quad (1)$$

where N is the particle number, \hat{q} is the unit vector in the direction of the scattering vector \vec{q} , $D_0 = \kappa_B T(6\pi\eta a)^{-1}$ is the Stokes-Einstein free-diffusion coefficient and \vec{r}^N represents the position of all colloidal particles. The brackets $\langle \dots \rangle$ stand for the ensemble average over all the spatial configurations of the particle systems.

Beenakker and Mazur have evaluated $H(q)$ applying a renormalization method valid at arbitrary volume fractions that has been successfully employed with hard sphere systems (Beenakker and Mazur 1984). This method has also been

used by Genz and Klein for dispersions of moderately charged particles (Genz and Klein 1991). The result is called the $\delta\gamma$ -expansion to zero order and can be expressed in terms of $S(q)$

$$H(q) = \frac{D_S(\phi)}{D_0} + \frac{3}{2\pi} \int_0^\infty d(aq') \left(\frac{\sin(aq')}{aq'} \right)^2 [1 + \phi S_{\gamma_0}(aq')]^{-1} \times \int_{-1}^1 dx (1 - x^2) (S(|q - q'|) - 1) \quad (2)$$

where $D_S(\phi)$ is the self-diffusion coefficient and $x = \cos(q', q)$. The function S_{γ_0} depends on the volume fraction ϕ through the scalars $\{\gamma_0^{(1)}\}$. An alternative hydrodynamic theory used for charged systems is the Pairwise Additivity Approximation. In our case we choose the Beenaker and Mazur formalism since the Pairwise Additivity Approximation needs as an input function in the calculations the radial distribution function which is described in the real space, and our experimental data, $S(q)$ are in the reciprocal one.

3 Experimental

Liposomes are composed of egg phosphatidylcholine (PC) and phosphatidylserine (PS) at a ratio of PS/PC = 0.25. PC is a zwitterionic phospholipid, comprising a choline and a phosphate group. At the working conditions we presume PC to be uncharged. By contrast, PS is an ionic phospholipid with an expected charge of $1e^-$ per molecule, which sets the total surface charge of our liposomes, thereby controlling the long-range repulsion between liposomes. The preparation technique used (Haropérez et al. 2003) leads to unilamellar vesicles that are rather monodisperse in size, which we determine by dynamic light scattering as 98 ± 5 nm. The volume fraction of the stock suspension was estimated from the lipid weight fraction, $30 \mu\text{mol/mL}$, and the mean outer and inner vesicle radii (a and b , respectively) (Stuchly et al. 1988), considering that the thickness $\Delta = a - b$ and the density of the phospholipid shell are 4.5 nm and 1.015 g/cm^3 , respectively (Huang and Mason 1978; Lasic 1991).

We perform static and dynamic light scattering using an ALV-5000 spectrophotometer with an Argon-Ion Laser (Coherent, model Innova 308) of wavelength $\lambda_0 = 514.5$ nm. Average intensities are obtained from several measurements at each angle, with different cell positions to minimize the effect of scratches on the cell surface. The intermediate scattering functions, $g_2(q, t)$, are recorded during a time around four orders of magnitude the decay time of the correlation function, which depends on density and angle, in order to obtain a good statistic. For ergodic systems, where the ensemble average of the intensity can be identified with the time average intensity, the field correlation function is obtained from the time correlation function of the intensity by the Siegert relation (Berne 1976) as

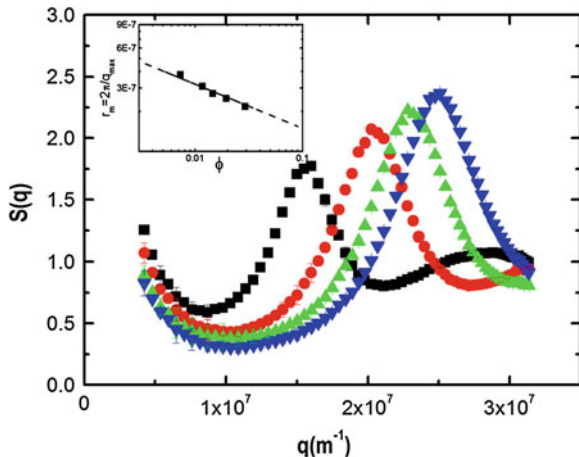


Fig. 1 Static structure factor for liposome dispersions as a function of wave vector at different volume fractions: 0.7, 1.16, 1.45, 1.93 %, (from *left* to *right*). Inset: Mean interparticle distance versus volume fraction. Experimental data (*squares*), theoretical values $0.9 \cdot n^{-1/3}$ (*dashed line*)

$$g_2(q, t) = \frac{\langle I(q, 0)I(q, t) \rangle_E}{\langle I(q, 0) \rangle_E^2} = 1 + |\beta g_1(q, t)|^2 \quad (3)$$

where $I(q, t)$ is the scattered intensity by the sample at time t and at scattering vector q and β is an experimental constant smaller than one. The ergodicity theorem allows replacing the ensemble average of the intensity, $I^E = \langle \dots \rangle_E$, by its time average, I^t , which is the magnitude that a correlator in a light scattering device provides.

As multiple scattering is strongly dependent on the particle concentration and particle refraction index, usually researchers eliminate it reducing the contrast between solvent and particles, reason why the medium composition has to be altered. In this case, the contrast of the system is intrinsically low since the refraction index of the liposomes, $n_l = 1.36$, is quite close to that of the water, $n_w = 1.33$ (Haro-Pérez et al. 2009).

Subsequently, several structures were formed for increasing liposome concentrations from 0.7 to 1.9% volume fraction. The suspensions studied in this work were kept for 5 days over a bed of ion exchange resin (Amberlite NRM-150).

4 Results

The static structure factor may be calculated by $S(q) = (\rho_0/\rho)(I(q)/I_0(q))$, where $I_0(q)$ is the light intensity scattered by a sample of non-interacting particles with number density ρ_0 . The results obtained are shown in Fig. 1. Obviously, this figure reveals the existence of liquid-like order in the vesicle suspension.

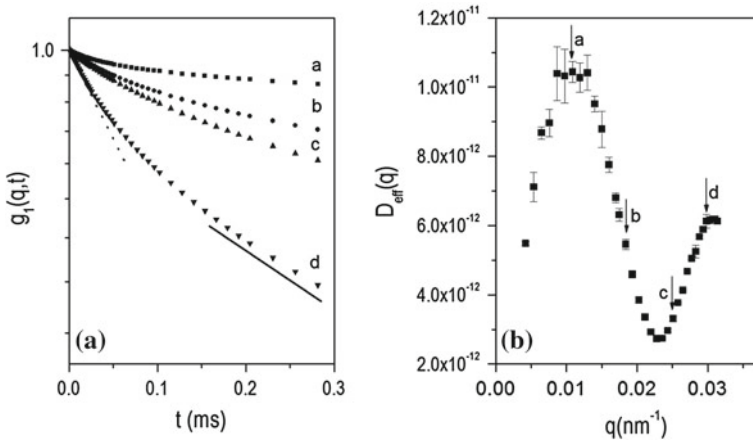


Fig. 2 **a** Field autocorrelation functions for a suspension at concentration $\phi = 1.45\%$ at the wave vectors marked with arrows in the *right side*: (a) 39° , (b) 69° , (c) 101° , (d) 133° . The *straight lines* are fits to the linear part related to the short-time dynamics (*dotted line*) and to the long-time dynamics (*solid line*). **b** Angular dependence of the effective diffusion coefficient for the same sample

The structure factor exhibits a pronounced maximum, whose position strongly depends on the particle volume fraction. The height and position of the main peak exhibit the expected behavior as the particle concentration increases, i.e., the peak height increases and their position shifts to large q -values as a function of $n^{-1/3}$, where n is the particle number density. This trend is characteristic of charged colloidal systems. However, we have seen in previous studies that the structure factor calculated theoretically by solving the Ornstein-Zernike equations along with a closure relation by assuming a purely repulsive potential can only describe the main peak of $S(q)$. The fits start to fail at lower q -values where we find an upturn of the static structure factor, being $S(q)$ larger than predicted. In a similar system, we could describe the low- q regime by assuming a mixed potential that comprises a long range repulsion and a shorter range attraction (Haro-Pérez et al. 2009).

Concerning the dynamic properties, dynamic light scattering measurements were performed via the *normalized* intensity correlation function. The analysis of the curves $\sqrt{g_2(q,t) - 1}$ versus t reveals interactions between liposomes have a marked effect: $\sqrt{g_2(q,t) - 1}$ departs strongly from a simple dependence on time and its decay cannot be described by a single exponential law as can be seen in Fig. 2a). In this plot several field correlation functions measured at different wave vectors for a given sample are displayed. In all cases, we observe that the decay of $g_1(q,t)$ is linear for $t \rightarrow 0$, so the short-time effective-diffusion coefficient can be measured from their slope $-D_{\text{eff}}q^2$ (Snook and Tough 1983), $g_1(q,t) = \exp(-q^2 D_{\text{eff}} t)$. To obtain D_{eff} , a cumulant expansion of second order was fitted to the autocorrelation function. The second linear regime found at longer times is related to the long-time dynamics, which is not the focus of the present work. The angular dependence of the normalized short-time effective diffusion coefficient D_{eff} on the wave vector is

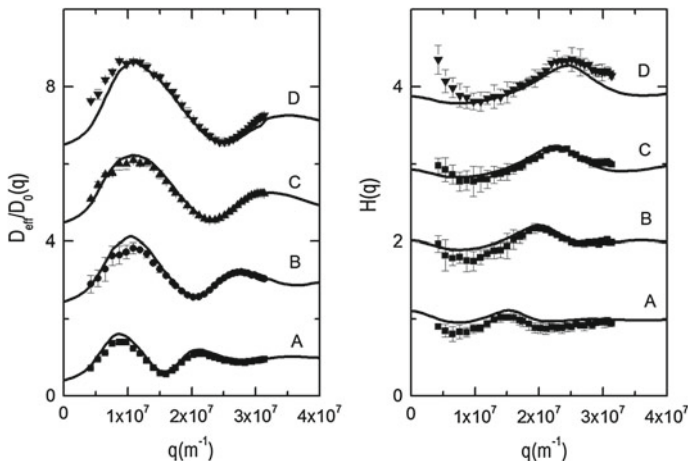


Fig. 3 *Left* Measured normalized effective diffusion coefficient at different volume fractions (a) 0.7 %, (b) 1.16 %, (c) 1.45 %, (d) 1.93 %, along with their corresponding theoretical fits according to $\delta\gamma$ expansion theory but with the experimental data of $S(q)$ used as input functions in the calculation of $H(q)$. Offset by 2. *Right* Comparison of their corresponding experimental hydrodynamic function at the same volume fractions (*solid squares*) with the theoretical fits computed according to $\delta\gamma$ expansion theory but with the experimental $S(q)$ used as input function in the calculation of $H(q)$ (*solid line*). Offset by 1

shown in Fig. 2b). These values are obtained by fitting the initial slope of the field correlation functions, as shown in Fig. 2a).

The single particle diffusion coefficient is measured by dynamic light scattering using diluted samples of liposomes where the measurements are performed at several angles. The obtained result was $D_0 = (4.98 \pm 0.20) \cdot 10^{-12} m^2/s$. Similarly, we analyze the rest of the samples. The angular behavior of the normalized effective diffusion coefficient for different volume fractions is plotted in Fig. 3 (left). We can observe the inverse of the curves show surprising similarities with their corresponding $S(q)$: an atypical upturn at the collective regime, a maximum occurring at the same scattering vector q_{max} where the static structure factor $S(q)$ has its first peak and the oscillatory behavior for $q > q_{max}$. The decrease at low q values of the normalized diffusion coefficient reflects that the dynamics in the collective regime slows down. Moreover, the minimum related to the mean peak of $S(q)$ shifts with concentration to larger q values in the same way $S(q)$ does. This link between structure and dynamics is well known (Ackerson 1976). A striking feature observed in the diffusive behavior of these dispersions is the appearance of a maximum in the collective regime, placed at the same position as the minimum of the static structure factor. This maximum occurs at a scale corresponding to 2–2.5 interparticle distance units, suggesting the existence of dynamical heterogeneities of these sizes.

Combining the effective diffusion coefficient $D_{eff}(q)$ and the measured static structure factor enables to determine the hydrodynamic function without taking recourse to any theoretical model beforehand. In Fig. 3 on the right, we plot $H(q)$ versus

q obtained for different volume fractions, whose structure factors are shown in Fig. 1. All the curves show common features: An increase at low- q values followed by a minima resembling the one of $S(q)$ and the presence of a pronounced peak (concentration dependent) at the same position as the particle-particle correlation peak of the static structure factor. The height and the width of the main peak increase with ϕ , and its value is always bigger than unity. This feature indicates hydrodynamic interactions speed up the system dynamics in the vicinity of the particle correlation peak, behaviour that has been previously observed in charged systems (Härtl et al. 1999; Rojas et al. 2003). As different authors reveal, this contrasts with hard-sphere dispersions, where the peak never exceeds one and its height decreases with increasing ϕ (Segré et al. 1995).

With regard to the behaviour at low q , we observed an unusual minimum that mimics the one observed in the structure factor. For a more detailed analysis, we perform a theoretical approach to explain our experimental results. As the particles we are dealing with are charged, we tried to fit the experimental $H(q)$ according to the Modified Beenakker and Mazur Formalism where the input parameter is the structure factor. In Fig. 3, comparisons of the experimental results to model calculations are presented. Firstly we tried to calculate the $H(q)$ by assuming a pure repulsive interaction potential to generate the input function $S(q)$, data are not shown. In this case the fits are rather poor, particularly at low wave vectors. This finding is predictable, as the statics could not be explained assuming a purely repulsive interaction potential. To describe our experiments we have used a different approach, instead of using as input the theoretical curves of $S(q)$, which failed as well at low angles to describe the static experiments, we employ the experimental $S(q)$ to evaluate the hydrodynamic function. In this case, we observe that the theory (solid lines) reproduces the tendency of the experimental curves at low wave vectors although there is a little vertical displacement. The error bars are considerable since the experimental values of the hydrodynamic function are obtained from the multiplication of two experimentally determined quantities. That is the reason why we prefer to compare the theoretical fits to the experiments in terms of the effective diffusion coefficient, where we can see a better agreement between experiments and theory, see solid lines in Fig. 3 (left). As we can see, the theory starts to fail for the most concentrated sample mainly at low wave vectors. These discrepancies could be due to the proximity of a phase transition where the theory would not be valid. The novelty of our findings is that the Beenakker-Mazur formalism, in principle devised to describe hydrodynamic interactions in hard spheres systems, and later adapted to charged particles, has now been applied to systems that show hallmarks of attraction. Moreover this theory provides a qualitative description of the experimental hydrodynamic functions.

5 Conclusions

In this work ordered colloidal suspensions from moderate to large volume fractions of charged liposomes are formed and their static and dynamic properties are analyzed by light scattering techniques. The particle-particle peak of the static structure factors

follows the usual $n^{-1/3}$ law, but there is an upraise at low q 's that cannot be explained by a purely repulsive interaction potential. Concerning the short-time dynamics, despite the lack of understanding of the $S(q)$ we can simply predict the short time dynamics by using the modified Beenaker-Mazur formalism, which describes the experimental data reasonably well. We have found the Beenaker-Mazur formalism is more general than expected and can be applied successfully to other kind of systems, not only repulsive, but also with attraction as long as the structural properties of the dispersion are well determined and used as input functions in the theory.

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References

- Ackerson BJ (1976) *J. Chem. Phys.* 64:242–246
- Beenakker CWJ, Mazur P (1984) *Physica A* 126:349–370
- Berne BJ, Pecora R (1976) *Dynamic light scattering: with applications to biology, chemistry and physics.* Wiley, New York
- Genz U, Klein R (1991) *Physica A* 171:26–42
- Haro-Pérez C, Quesada-Pérez M, Callejas-Fernández J, Casals E, Estelrich J, Hidalgo-Álvarez R (2003) *J. Chem. Phys.* 118:5173–6167
- Haro-Pérez C, Rojas-Ochoa LF, Trappe V, Castañeda-Priego R, Estelrich J, Quesada-Pérez M, Callejas-Fernández J, Hidalgo-Álvarez R (2009) *Structural and functional properties of colloidal systems*, vol. 146. CRC Press, Taylor and Francis Group, London, pp 77–91
- Haro-Pérez C, Rojas-Ochoa LF, Castañeda-Priego R, Quesada-Pérez M, Callejas-Fernández J, Hidalgo-Álvarez R, Trappe V (2009) *Phys. Rev. Lett.* 102(081301):1–4
- Härtl W, Beck C, Hempelmann R (1999) *J. Chem. Phys.* 110:7070–7072
- Huang C, Mason JF (1978) *Pro. Natl. Acad. Sci. U.S.A.* 75:308–310
- Lasic DD (1991) *Liposomes: from physics to applications.* Elsevier, Amsterdam
- Phalakornkul JK, Gast AP, Pecora R, Nägele G, Ferrante A, Mandl-Steininger B, Klein R (1996) *Phys. Rev. E* 54:661–675
- Philipse AP, Vrij A (1988) *J. Chem. Phys.* 88:6459–6470
- Pusey PN, Segrè PN, Behrend OP, Meeker SP, Poon WCK (1997) *Physica A* 235:1–8
- Pusey PN (1991) *Liquids, freezing and glass transition.* In: Hansen JP, Levesque D, Zinn-Justin J (eds) *Volume Session L1 of Les Houches.* Elsevier Science Publications, Les Houches, pp 763–942
- Riese DO, Ewgdam GH, Vos WL, Sprik R, Fenistein D, Bongaerts JHH, Grübel G (2000) *Phys. Rev. Lett.* 85:5460–5463
- Rojas LF, Vavrin R, Urban C, Kohlbrecher J, Stradner A, Scheffold F, Schurtenberger P (2003) *Faraday Discuss.* 123:385–400
- Segré PN, Behrend OP, Pusey PN (1995) *Phys. Rev. E* 52:5070–5083
- Snook I, Tough RJA (1983) *J. Chem. Phys.* 78:5825–5836
- Stuchly MA, Stuchly SS, Liburdy RP, Rousseau DA (1988) *Phys. Med. Biol.* 33:1309–1324