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Determination of particle concentration in latexes by turbidimetry

J.M. Irache, C. Durrer, G. Ponchel and D. Duchêne

Laboratoire de Pharmacie Galénique et de Biopharmacie, URA CNRS 1218, Université Paris-Sud, 5, rue Jean-Baptiste Clément, 92296 Châtenay-Malabry (France)

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Summary

The capacities and limits of turbidimetry for determining the particle concentration in latexes were studied. The size dependence of the specific turbidity (τ/C) was demonstrated for monodisperse latexes with sizes between 114 and 1060 nm. The particle concentration (C) of different latexes was calculated from turbidity values. The observations were also valid for polydisperse systems and the comparison between turbidimetric and gravimetric concentrations showed similar results ($p < 0.05$). By turbidimetry the exact concentration of even very small samples could be determined in a non-destructive way.

Latexes such as nanoparticulate or nanocapsule systems are not only used for agglutination tests in diagnostic studies (Manil et al., 1986) but are also a subject of great interest for pharmaceutical applications (Guiot and Couvreuer, 1986; Kreuter, 1991). One of the most common practical problems proper to these new carrier systems is to determine their concentration in rather small samples or diluted samples after operations like filtration, dialysis or protein coupling.

The turbidity τ of a colloidal suspension is the reduction in intensity of an incident beam caused by light scattering. For particles with a diameter near the wavelength of the light, Mie's theory of light scattering in presence of monodisperse spherical particles can be applied (Mie, 1908); for a monodisperse suspension, the following equations are valid (Wales, 1962; Kourti and MacGregor, 1991):

$$\tau = (1/l)\ \ln(I_o/I) = \pi K D^2 N/4 \qquad (1)$$

where l is the length of scattering path, I_o and I denote the incident and emergent light intensities, D is the particle diameter and N is the number of particles per volume unit. The scattering coefficient K is a function of two parameters α and m, where α is the relative size of the particle to the wavelength of the light in the medium and m the ratio of the refractive index of the particles to that of the medium.

$$\alpha = \pi n_o D/\lambda \qquad (2)$$

$$m = n/n_o \qquad (3)$$

Correspondence to: G. Ponchel, Laboratoire de Pharmacie Galénique et de Biopharmacie, URA CNRS 1218, Université Paris-Sud, 5, rue Jean-Baptiste Clément, 92296 Châtenay-Malabry, France.

One of the most widely used turbidimetric techniques consists in the determination of the specific turbidity (τ/C). For a monodisperse latex suspension τ/C can be written as (Yang and Hojg, 1979):

$$(\tau/C) = [3/2K(\alpha,m)]/\rho D \qquad (4)$$

where C is the latex concentration expressed as mass of polymer per volume and ρ is the density of polymer.

The aim of this work was to study the capacities and limits of specific turbidity as a simple technique for determining particle concentration in latexes.

Poly(styrene) latex beads CLB-9, LB-1, LB-3 and LB-5 were purchased from Sigma (St. Quentin-Fallavier, France); CML-005, CML-009, PSL-221, PSL-228, PSL-229 and PSL-213 from Polymer Laboratories Ltd (Church Stretton, U.K.), Polybead® Carboxylate Microspheres (PCM) of 0.20, 0.75 and 1.0 μm and Polybead® Amino Microspheres (PAM) of 0.50, 0.75 and 1.0 μm from Polysciences Inc. (Eppelheim, Germany), and latex microspheres (LM) of 0.80 μm from Coultronics (Margery, France). Dilutions of the latexes were made in order to measure turbidity values between 0.1 and 1 unit. For each latex, turbidity was measured at 300, 550, and 750 nm wavelengths on a Perkin Elmer UV-Vis Lambda 5 spectrophotometer. The lower limit of λ was set by the polymer absorption at around 260 nm and the upper limit was fixed at 750 nm due to the instrumental restrictions. Quartz cuvets with an optical path of 1 cm were used.

The concentrations (mass of solids/volume) of the following latexes were analysed by gravimetry: PSL-228, LM-0.80, PSL-213, PSL-229 and CML-009. For this purpose a Mettler AJ100 analytical balance was used (Mettler-Toledo AG, Switzerland, $d = 0.1$ mg)

To confirm the size data given by the manufacturers, dilute suspensions of the latexes were measured by photon correlation spectroscopy (PCS) on a Coulter® N4MD submicron particle analyzer (Coultronics, Margency, France).

It should be pointed out here that all the latexes used in this study were suspensions of spherical, monodisperse and non-absorbing undyed particles. The main characteristics of the different latexes and the weight diameters (D_w) obtained by PCS are shown in Table 1. The diameter values from turbidimetry were assigned to the weight diameter (D_w) because it is generally the only value which is valid for different methods of size determination (Goulari et al., 1987; Kourti and MacGregor, 1991).

TABLE 1

Latexes studied and their characteristics

Product	Active group	TEM diameter (nm) [a] (±SD)	Weight diameter (nm) [b] (±SD)	Content of solids [a] (%)
LB-1	–	114±20	108±16	10
PSL-213	–	–	209±15	–
PCM-0.2	COOH	210± 6	230±13	2.5
LB-3	–	303± 4	306±16	10
CML-005	COOH	–	320±26	28
PAM-0.5	NH_2	470± 5	457±28	2.5
LB-5	–	460± 5	466±35	10
PSL-228	–	–	626±53	–
CML-009	COOH	–	654±42	–
PSL-221	–	–	670±80	11
PCM-0.75	COOH	790±34	711±24	2.5
PSL-229	–	–	760±96	–
LM-0.80	–	–	804±31	–
PAM-0.75	NH_2	750± 6	805±51	2.5
PCM-1.0	COOH	880± 9	875±40	2.5
CLB-9	COOH	885±38	890±45	10
PAM-1.0	NH_2	1000± 9	1060±52	2.5

[a] Data obtained from supplier.

[b] Data obtained from photon correlation spectroscopy (PCS).

Standard curves of different latexes were established by referring the specific turbidity (τ/C) to the weight diameter (Fig. 1). This was made at $\lambda = 750$ nm for large particles (D_w between 180 and 1325 nm), at $\lambda = 550$ nm for intermediate particles (D_w between 130 and 970 nm) and at $\lambda = 300$ nm for small particles (D_w between 70 and 530 nm), in order to respect validity limits for α ($1 < \alpha < 7.4$), according to previous specifications given by other authors (Cheesman, 1978; Kourti and MacGregor, 1991). In all cases a good linear relationship between specific turbidity and D_w (PCS) was observed, i.e., high correlation coefficients and small slope errors (data not shown).

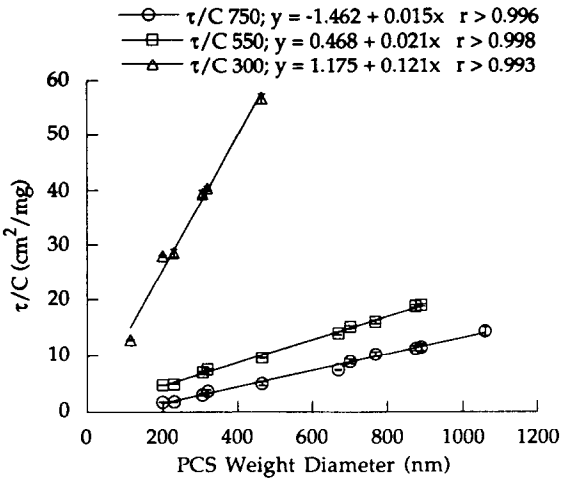

Fig. 1. Dependence of the specific turbidity (τ/C) on the particle size at λ = 750 (○), 550 (□) and 300 nm (△).

In fact, specific turbidity is one of the most common methods of determining the particle size as shown for latexes of poly(styrene) (Yang and Hojg, 1979; Kourti et al., 1990) and poly(vinyl acetate) (Zollars, 1980), but the concentration of the colloidal suspension must be known for this. In the present case, the concentration determination requires only the knowledge of the particle size, obtained by any available method, and establishing a standard curve.

Furthermore, five different monodisperse latexes which were tested by turbidimetry and gravimetry as the method of reference (Table 2. In all cases, the differences between both methods were not significant ($p < 0.05$), but large amounts of samples were necessary to obtain accurate values of concentration by gravimetry.

TABLE 2

Comparison of concentrations obtained by turbidimetry (average of three replicates) and gravimetry (average of two replicates)

Latexes	D_w (nm)	Turbidimetry concentration (mg/ml) (±SD)	Gravimetry concentration (mg/ml) (±SD)
PSL-228	626±53	95.11±2.98 (0.1) [a]	97.53±3.12 (1) [a]
LM-0.80	804±31	103.62±1.54 (0.1) [a]	105.34±1.87 (1) [a]
PSL-213	209±15	103.05±2.37 (0.1) [a]	101.75±3.57 (1) [a]
PSL-229	760±96	24.01±0.80 (0.1) [a]	23.11±0.64 (3) [a]
CML-009	654±42	47.50±1.12 (0.1) [a]	46.78±1.95 (1.5) [a]

[a] Volume (ml) of latex sample used.

TABLE 3

Particle concentrations (average of three replicates) of different mixtures of two latexes; theoretical concentration of the mixture: 25 μg/ml (data based on supplier information)

Mixture of		D_w (nm) (±SD)	Experimental concentration (μg/ml) (±SD)	Accuracy (%)
LCM-0.2 (%)	LCM-1.0 (%)			
20	80	808±409	23.62±1.03	−5.52
40	60	640±287	24.24±0.89	−3.04
60	40	506±217	23.86±0.95	−4.56
80	80	348±182	25.93±1.17	3.72

Finally, the behaviour of polydisperse latexes was investigated. Some 25 μg/ml mixtures of the latexes PCM-0.2 (D_w = 230 ± 13 nm) and PCM-1.0 (D_w = 875 ± 40 nm) were analyzed in order to test the possibility to determine the concentration of polydisperse latexes based on the average weight diameter obtained from PCS. As shown in Table 3, differences between experimental and theoretical concentrations were around 5%. These results suggest that polydisperse systems can be succesfully investigated by the turbidimetric method.

Despite some disadvantages (turbidimetry is a complex theory and the refractive index of the particles must be known to estimate α), the main advantages of the turbidity method described are its rapidity, low price and simplicity. Further, the measurements do not perturb the system under investigation, and it is possible to determine the accurate concentration with very small amounts of samples compared to other methods.

References

Cheesman, G.C.N., A rapid method for the measurement of the average particle size of non-uniform lattices and dispersions. In Groves, M.J. (Ed.), *Particle Size Analysis*, Heyden, Norfolk, 1978, pp. 160–166.

Goulari, E., Bazzi, G., Goulari, E. and Annapragade, A., Latex particle size distribution from multiwavelength turbidity spectra. *Part. Charact.*, 4 (1987) 96–100.

Guiot, P. and Couvreur, P., *Polymeric Nanoparticles and Microspheres*, CRC Press, Boca Raton, FL, 1986.

Kourti, T., MacGregor, J.F. and Hamielec, A.E., Particle size determination using turbidimetry. Capabilities, limitations and evaluation for on-line applications. *Polym. Mater. Sci. Eng.,* 62 (1990) 301–305.

Kourti, T. and MacGregor, J.F., Particle size determination using turbidimetry. Capabilities, limitations and evaluation for on-line applications, in particle size distribution. II. *ACS Symp. Ser.,* 1991, pp. 34–63.

Kreuter, J., Peroral administration of nanoparticles. *Adv. Drug Del. Rev.,* 7 (1991) 71–86.

Manil, L., Roblot-Treupel, L. and Couvreur, P., Isobutyl cyanoacrylate nanoparticles as a solid phase for an efficient immunoradiometric assay. *Biomaterials,* 7 (1986) 212–216.

Mie, G., Beiträge zur Optik trüber Medien, speziell kolloidaler Metallösungen. *Ann. Phys.,* 25 (1978) 377–445.

Wales, M., Particle size distribution in rubber latex. *J. Phys. Chem.,* 66 (1962) 1768–1772.

Yang, K.C. and Hojg, R., Estimation of particle size distributions from turbidimetric measurements. *Anal. Chem.,* 51 (1979) 758–763.

Zollars, R.L., Turbidimetric method for on-line determination of latex particle number and particle size distribution. *J. Colloid Interface Sci.,* 74 (1980) 163–172.