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Hand-Held Injection Market Drivers

IN THIS ISSUE



INTERVIEW WITH
MACULEAR'S
CEO

PHILIP RALSTON

**Beyond The
Headlines** 18

Derek Hennecke

**Functional
Excipients** 22

João Correia
António Bica

**Patient
Compliance** 30

Steven Hamlen, MBA
Karen MacGregor, PhD

**Zeta
Potential 2** 50

David Fairhurst, PhD
Robert W. Lee, PhD

FEATURING

SPECIALTY 
Strategies For
Business Development **PHARMA**

**Outsourcing
Trends** 64

Jennifer Brice
Barath Shankar

**Development
Strategies** 66

Andrew MacGarvey

The science & business of drug development in specialty pharma, biotechnology, and drug delivery



**Fethi Trabelsi,
PhD**

Importance of
Pharmacokinetic/
Pharmacodynamic
Studies During
Biosimilar Clinical
Development



Cindy Dubin

Biologics, Self-
Administration &
Patient Adherence:
Creating a Lucrative
Hand-Held Injection
Market



**Josef Bossart,
PhD**

DDEP Sales &
Prescription
Performance 2010,
Hints of the Future

ZETA POTENTIAL

The Zeta Potential & its Use in Pharmaceutical Applications - Part 2: Measurement Techniques & Uses

By: David Fairhurst, PhD; Robert W. Lee, PhD

INTRODUCTION

In Part 1 (July 2011 issue) of this review, we covered charged interfaces (in both polar and non-polar media) and introduced the concept of the zeta potential (ZP). In this concluding Part 2, we discuss techniques to measure ZP and illustrate the utility of its measurement.

MEASUREMENT OF ZETA POTENTIAL: ELECTROKINETIC PHENOMENA

An electric double-layer (EDL) exists between a surface and solution. Any relative motion between the rigid and mobile parts of the EDL will result in the generation of an electrokinetic potential. There are four fundamental ways (Figure 1) that electrokinetic potential can be produced: electrophoresis, electro-osmosis, streaming potential, and sedimentation potential.¹ The parameter used to describe this real, measurable potential is the ZP, introduced in Part 1, in which the theoretical model for the EDL was discussed.

Measurements made using

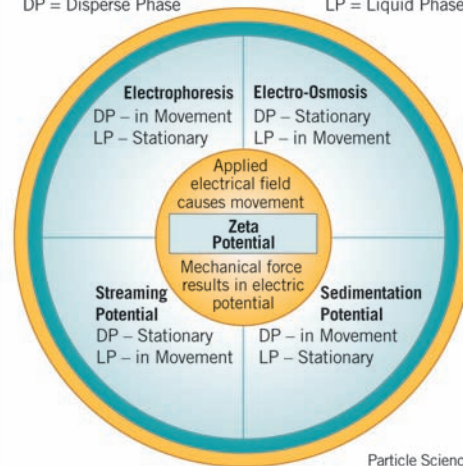
techniques based on the four effects should give the same value for ZP. However, owing to a variety of contributing factors, this is not necessarily the case. These include assumptions in the model for the EDL, inadequacies in the theory,

experimental limitations in the design and construction of instrumentation, and differences in sample preparation.² The choice of method should be based on the specific application under investigation.

FIGURE 1

RELATIONSHIP BETWEEN THE FOUR TYPES OF ELECTROKINETIC PHENOMENA

DP = Disperse Phase LP = Liquid Phase



Particle Sciences

TABLE 1

Material	ZP (mV)
O/W Emulsions	>15
Polymer Latices	>20
Metal Oxides	>40
Metal Sols	>70

Rule of Thumb for Electrostatic Stabilization

Particle Electrophoresis

The most common technique based on microelectrophoresis is electrophoretic light scattering (ELS), sometimes called laser-Doppler electrophoresis (LDE). It is fast, and the latest commercial devices are fully automated, requiring little input from the user. ELS/LDE instruments yield a histogram of the ZP of the particles within the sample suspension, and most devices also measure the particle size distribution.

ZETA POTENTIAL REFERENCE MATERIALS

There is no accepted universal standard for ZP measurements. However, there are a number of reference materials supplied by the various instrument manufacturers. These are well-characterized suspensions whose ZP has been established over hundreds of measurements over many years; it is typically some stated value for ZP ± 10% of that value.

The nearest “official” reference material is a sample of Goethite, α-FeO(OH), manufactured and distributed by NIST (Gaithersburg, MD), which when prepared under standard conditions, has a certified electrophoretic mobility value of +2.53 x

10⁻⁸ ± 0.12 x 10⁻⁸ m²V⁻¹s⁻¹, equivalent to a ZP of +32.5 mV ± 0.12 mV.³

Unfortunately, the Goethite suspension has to be freshly prepared before each use, and the protocol is not without travail. Further, being positively charged, the Goethite particles tend to coat the walls and surfaces of an instrument measurement cell; clean-up can be very tedious.

SYSTEMS HAVING VERY SMALL ZETA POTENTIALS

The ELS method works very well for aqueous systems in which the solution ionic conductance is moderate. However, human red blood cells (RBC) and microorganisms, such as bacteria and viruses, are suspended in water under physiological (isotonic) conditions (0.145 M NaCl, pH 6.8) in which the ionic strength and conductance are high.^{4,5} As the electrolyte concentration increases, the value of ZP falls owing to the shielding effect of the ion atmosphere around the particle (see Part 1, July 2011). Consequently, the ZP of RBC in isotonic solution is almost an order of magnitude smaller than in distilled water.^{5,6} Some body fluids can exceed the isotonic ionic strength by an order of magnitude, and salt levels in environmental systems, such as brine and sea

water, can be very high (~ 2 M).⁷ Obtaining reliable data in when the solution conductivity is so large poses a challenge for ELS/LDE devices because of Joule heating and other undesirable effects.^{8,9}

Particles that are sterically stabilized by adsorbed non-ionic surfactants, macromolecules, and synthetic polymers all have ZP values at or near zero; making reliable, reproducible measurements on such systems almost impossible.²

Non-aqueous systems also pose a challenge because the viscosities of many organic liquids are higher than water, and the dielectric constants are invariably lower. For example, PEG 200 (at RT) has a viscosity of ~ 100 cP and a dielectric of ~ 12; ZP measurements by ELS in this fluid would be impossible.

Phase Analysis Light Scattering

The aforementioned difficulties can be overcome by using phase modulation in place of frequency spectrum analysis.^{10,11} Termed phase analysis light scattering

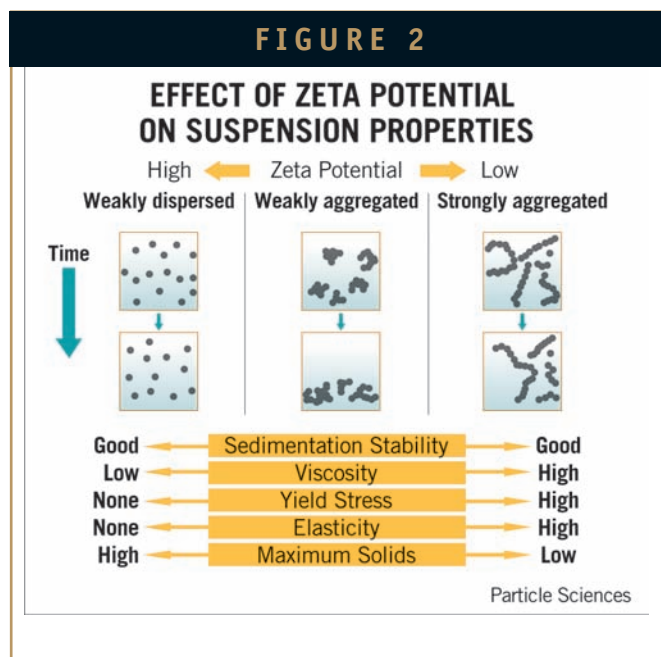
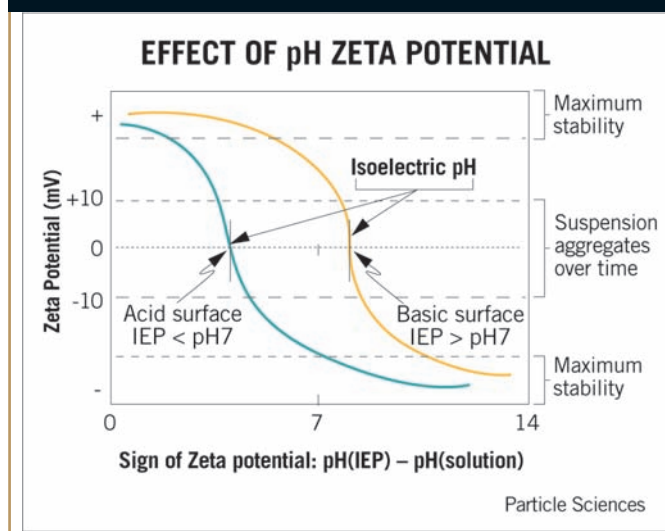


FIGURE 3



(PALS), this methodology has much superior sensitivity compared to conventional ELS/LDE. At 37°C, the viscosity of PEG 200 is ~ 30 cP, and a sample of casein dispersed in it has a measured ZP of ca -0.3 mV in contrast to a typical literature value for casein in water of ca -75mV.¹²

The limitation of the PALS method is that it will only give an average value for ZP, ie, there is no histogram. Hence, it cannot be used for systems in which mixtures of materials are used.

CONCENTRATES & OTHER UNUSUAL DISPERSIONS

Instrumentation based on ELS/LDE or PALS require that the system under measurement be initially diluted, but there is always a potential concern that the dilution process can produce instability; one classic example is emulsions. Another issue is the subsequent extrapolation of data from exceptionally dilute suspensions to the practical relevant concentration of the formulation. For biological systems,

the problem of small amounts of contaminants in dilute systems can be overwhelming, whereas in concentrates, this can be almost ignored. Finally, ELS and PALS are light scattering techniques and so have limitations with opaque (ie, light

absorbing) materials (ie, carbon black) or light sensitive materials (ie, selenium or silver compounds) or systems in which the particles have motility (ie, sperm).

Electroacoustic Attenuation

In sedimentation potential measurements (Figure 1), the disperse phase moves through a stationary liquid under the influence of gravity (the mechanical force) to produce an electric potential.¹³ The gravitational force can be replaced by an acoustic field to give an AC equivalent of the DC sedimentation potential. Termed electroacoustic attenuation (EAA), there are two principal methods: colloid vibration potential (CVP) and electrokinetic sonic amplitude (ESA); a comprehensive review of ultrasound techniques has been made by Dukhin.¹⁴⁻¹⁷

A major advantage of EAA is that the sample under investigation need not be stationary, a necessary requirement of ELS and PALS instrumentation. The prime disadvantage of the EAA technique is interference from the presence of air

bubbles.¹⁷ The lower limit of concentration that can be reliably measured is about 1%, and at high concentrations (> 50%), technical issues limit its usefulness.

It should not be expected that the value of ZP determined at high concentration using EAA should be the same as that determined under dilute concentrations using ELS or PALS. At very high solids concentration, interpretation of experimental measurements is complicated because of possible EDL overlap.¹⁸

Thus, if a suspension is prepared at one concentration but used at some other lower concentration, it is critical that measurements be made at the use concentration; measurements of ZP as a function of suspension concentration can often be illuminating.

MASSIVE & IRREGULAR SHAPE SOLID MATERIALS

There are applications in which the surface under investigation is not a particle. Examples include hair, bone, skin, polymer films, fibers, membranes, filter and porous materials, paper pulp, and metal foil. In addition, coarse and massive solids that are of irregular shape are simply not amenable because of their physical dimensions to be measured by ELS/PALS and EAA techniques.

Streaming Potential

Measurements on such systems can best be made using the streaming potential (SP) method.¹⁹ A major advantage of the

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SP technique is the ability to investigate adsorption/desorption phenomena *in situ*; the flow-through nature of the method makes it ideal for the study of long-term processes. However, there are very few commercial manufacturers of SP instrumentation.

THE USEFULNESS OF ZETA POTENTIAL MEASUREMENTS

The Relation of Zeta Potential to Suspension Behavior

All suspensions are inherently thermodynamically unstable and will over time, through random motion of particles, aggregate unless sufficient repulsive forces are present. The repulsive force between

particles, V_R , which keeps particles from aggregating, is illustrated in the following equation:

Equation 1.

$$V_R = D a \psi_d^2 \text{ [Geometric term]}$$

Where, D is a dimensionless constant related to the permittivity (dielectric constant) of the suspension medium, a is the particle size, and ψ_d is the Stern potential. The Geometric term is a function of particle radius, the Debye length, and the distance of separation between particles.

ZP is related to surface charge and can be substituted for the Stern potential,

ψ_d . Thus, we can see that for a fixed medium, particle size and ionic strength, V_R will increase as ZP gets larger. Hence, a widely used application of ZP is to monitor the stability of electrostatic suspensions. It is particularly useful to predict the resistance of such dispersions to coagulation by electrolytes, by determining the critical ZP, ie, the value of ZP below which the suspension is coagulated. Table 1 lists the approximate minimum values of ZP needed to electrostatically stabilize materials of different dielectric constant in neutral (ie, pH 7 at 25°C) water.

The values of ZP in Table 1 are meant as guides only. APIs can be considered similar (in this regard) to polymer latex

TABLE 2

Material	pH Value of IEP
Oxides	
Silicon Dioxide	2
Titanium Dioxide (Anatase)	4
Titanium Dioxide (Rutile)	6
Iron Oxide	8
Aluminum Oxide	9
Zinc Oxide	10
Magnesium Oxide	12
Proteins	
Ovalbumin	4.0
Casein	4.6
Gelatin	4.8
Streptavin	5.0
BSA	5.0
Lactoglobulin	5.3
Rapeseed 12S	7.0
Ribonuclease	9.5
Avidin	10.5

Isoelectric Point of Materials

particles. ZP measurement is often used in determining the critical coagulation concentration (CCC) of an electrolyte (the minimum concentration required for the onset of coagulation); the CCC is proportional to ZP^4/z^2 (where z is the electrolyte counter-ion valence).

A variety of products require rheological control as an integral part of product requirements. Effective control of rheology relies on knowledge of the effect of formulation and process variables. Figure 2 illustrates the relation between ZP and the rheological characteristics of suspensions. A high ZP produces a well-dispersed suspension. This in turn results in good sedimentation stability and high solids loading capacity. This type of suspension will have fluid-like characteristics, ie, low viscosity, extremely low elasticity, and relatively linear

viscoelastic behavior as a function of shear or strain. In contrast, a low ZP results in particle association. The formation of a strong network structure provides good sedimentation stability but greatly reduced solids loading capacity. Here, the rheological behavior would be characterized by a high viscosity, a high elasticity, and a yield point at some critical strain.

in the range about ± 10 mV, a suspension will be unstable and will aggregate over time. If the IEP is known, then the sign of ZP can be determined from the solution pH.

The second is the magnitude of the ZP increases (either side of the IEP) but eventually plateaus. The condition of maximum dissociation of any surface functional groups and can be used to great effect when dispersing particles; the greater the magnitude of ZP, the less the need for any additional dispersing aid. For proteins and charged macromolecules, the ZP value at this plateau generally increases with increasing MW. For example, at pH 9, the ZP of different gelatins can range from as low as -15 mV to more than -50 mV. Materials that carry a negative charge can be made to be positive by adding PDI or CMA.

Finally, particles with an IEP < pH 7 have acidic character, while those with an IEP > pH 7 are basic. An example of this is carbon black. Acidic carbon blacks have an IEP in the pH range of 3 to 4, while basic carbon blacks have an IEP in the pH range of 8 to 9. Table 2 lists the typical IEP of some common oxides and proteins.

DETERMINATION OF THE ISO-ELECTRIC POINT OF MATERIALS

All charged materials will have an isoelectric point (IEP), defined as the condition when the ZP is zero. This is usually achieved by addition of potential determining ions (PDI), which for many materials, means H^+/OH^- (ie, a function of pH). It can also be attained by specific adsorption of charge modifying agents (CMA).

Figure 3 is a plot of ZP versus pH from which a number of important points can be noted. The first is that at the IEP, a particle suspension will have no resistance to aggregation. This is used in applications such as waste water treatment. If the ZP is

CONCLUSION

ZP measurement is a very useful but often under-utilized technique that can provide information about the material surface-solution interface. Knowledge of ZP can be used to predict and control the stability of suspensions and emulsions; measurement of ZP is often the key to

understanding dispersion and aggregation processes. The presence, or absence of charged groups/moieties on the surface of materials, as revealed by their ZP, can directly affect their performance and processing characteristics in suspension.

The sign and magnitude of ZP affects process control, quality control, and product specification; at the simplest level, it can help maintain a more consistent product and at a complex level, it can improve product quality and performance. ◆

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BIOGRAPHIES



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