

Protocol Zeta potential

1. Method

ZetaPALS measures zeta potential using Phase Analysis Light Scattering, to determine the electrophoretic mobility of charged, colloidal suspensions in an electric field. A few [mL] of suspension are required to carry out the measurements. The measurements are conducted in conductive liquids, such as KNO_3 , at different pH. Just a few minutes are required for the sample and cell to equilibrate with the actively controlled temperature environment inside the instrument. Please note that avoiding dust is essential to carry out these measurements. It is often necessary to filter the solution of dispersant used for preparing the various dilutions and to wash all vessels in order to avoid the presence of dust.

2. Equipment

- Measurement device: Brookhaven BI-9000AT (more info from <http://www.bic.com>)
- Software versions (zetapw32.exe: 3.57; bi_zetapw32.dll: 3.4; Eclectic: 4.4)
- A special electrode for zeta potential measurement:
 - o AQ-878 for SPION only
 - o AQ-1047 for the other suspensions
- Disposable, acrylic square cells (4 faces) are used for aqueous and simple alcohol suspensions;
- A special glass cell can be used with aggressive solvents;
- Ultrasonication bath in MXD 313

3. Cleaning of the electrodes

To do at the beginning of each new session and after each measurements.

- Rinse the electrode with ultra-pure water
- Deep the electrode in a beaker of ultra-pure water: **only the external electrodes parts should be in contact with water**
- Put the beaker in ultrasonic bath for 0.5 to 1 min
- Rinse the electrode with ultra-pure water
- Repeat these operations 2 times more
- Dry the electrodes with soft paper

4. Preparations of the samples

- The ideal concentration range for the measurement is 0.1-10 [mg]/[mL].
- Put your suspension in ultrasonic bath for 5-15 minutes
- Approximately 1.5 mL of this suspension is then transferred to the specific cell of measurement (4 faces).
- **If the sample is prepared in an aggressive solvent, use a specific glass electrode**
- Introduce the electrodes in the cell (they should be fully inserted), add it inside the ZetaPals.
- Connect it to the equipment. Close the door.
- It is recommended to perform 2 measurements per sample, at each pH (3, 6 and 9).

- It can take some time to reach the equilibrium state. The time of measurement with respect to the time of first contact with H₂O must be noted. If there is an obvious trend in the measurement the sample should be left to equilibrate for a given time (1 hour to 24 hours), and the measurement repeated.

5. Operations

- Switch on the instrument (using the main switch or behind instrument).
- Type the username: ZetaPals. There is no password.
- Click twice the icon "BIC Zeta Potential Analyser "
 - o One can hear a "clic" which indicates the laser has started
 - o Before starting a measurement a delay of minimum 5 minutes must be respected to stabilize the laser and the temperature in the cell
- Select the menu "File", then "Database", double click on the file name you want to start with, then select exit.
 - o If you don't have any user name you can add it on create folder.
- Click on "Clear" before starting a new measurement
- You can choose 3 different measurements methods on Mode in the software main window:
 - o Single cycle: You can choose a number of runs with 1 cycle (routine measurements)
 - o Multiple cycles: You can choose a number of runs and of cycles (for stable but difficult or low Zeta potential measurements)
 - o High precision: 30 measurements will be done and only the results with a deviation smaller than 2σ will be taken into account. You can't change the number of runs and cycles
- In the bottom of the ZetaPals main window, click on "Parameters", and fill the table as described with the example of SPION

Sample ID	SPION-MB19
Operator ID	LM
Notes	100 ug/mL ; NaCl 10 mM
Cycles	5 (between 1 to 5): Number of cycles in 1 run
Runs	10 (between 1 to 10)
Inter Cycle Delay	5 sec
Temperature	25°C (it is advised to work at room temperature to improve the sample stability, but this can be changed if necessary)
Liquid aqueous	(viscosity, refractive index are automatic) You can add your own liquid if it is not in the list.
Batch number	From 1 to 99999
pH	6
Concentration	100 ug/mL
Particle size	15 nm
User 1	You can add any information you want
User 2	You can add any information you want
Select Auto save results (Very important) <input checked="" type="checkbox"/>	

The information in the shaded grey area is not important for the measurements, only as description.

- Click on “Start”
- For the first measurement, the software will optimize the count rate.
- It is better to optimize the count rate for each measurement:
 - o Restart the software for each measurement
- During the measurement:
 - o Check “Reference count rate”: must be between 1500 and 2500 kcps, and stable
 - o The count rate must be around 500 kcps
 - o If it is not the case you should dilute or up-concentrate your sample.
- At the end of the measurement, it is possible to delete some runs which are far from the mean value of the zeta potential, by double clicking on them (Only for single and multiple cycle modes).
- A standard deviation of ± 5 [mV] is good. A standard deviation of ± 10 [mV] is acceptable.
- **Please fill in the ZetaPals notebook**

6. Presentation of the results, data storage and data treatment

Save your results in my_documents/my_folder. If you don't have any, please create one.

6.1. Print the results

It will create a Pdf report.

- File/Print File/Report Print Options: choose the options
- File/ Print report
- The PDF creator will open
- Choose the name of the report and click on “Enregistrer”
- Save your pdf file in my_documents/my_folder

6.2. Create a report

It will create a text file which could be import in Excel.

- File/Report File Settings: choose the options
- Create Report File From Saved Measurements
- Choose the measurement you want to create a report
- Save the report as text file (.txt) in my documents/my_folder

6.3. Create a graph file

It will create a text file of the Zeta average graph.

- Click on Zoom
- Click to Copy to Spreadsheet
- Paste in a text file
- Save the report as text file (.txt) in my documents/my_folder

If you click on “Autosave results”, all your results will be saved on the ZetaPals computer.