

HOW TO USE PHOTOPETTE®'S DISPOSABLE CUVETIP® CORRECTLY

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- This application note provides tips and recommendations for the correct use of the device and its disposable CuveTip®

OBJECTIVE

The application note provides tips and tricks on how to use the Photopette's CuveTip® correctly. It teaches the recommended workflow to perform a measurement and provides information on limitations of the CuveTip®. A general introduction to Photopette® can be found in the "Photopette® User Manual" and "Quick Start Guide" available at the Tip Biosystems webpage. [1, 2]

INTRODUCTION

Photopette® is a revolutionary first-of-its-kind handheld fixed wavelength spectrophotometer manufactured by Tip Biosystems Pte Ltd, Singapore. The Photopette® device is used with the disposable CuveTip® that replaces the classical cuvettes workflow. The system is easy to use by both technical and non-technical users for photometric measurements, and can boost productivity in laboratories or for field measurements. Users are advised to follow the following guidelines for the CuveTip® to achieve higher measurement accuracy with the Photopette®.

MATERIALS AND APPARATUS

Instruments

- Photopette® device
- CuveTips®

Reagents

- As per the requirements of the users' applications.

METHOD

In the following section, we introduce different methods and give tips on how to work with the Photopette's CuveTip® efficiently and to avoid errors.

WHAT IS THE WORKFLOW WITH THE CUVETIP?

We recommend to use the same CuveTip® for auto zero and the sample measurement. This will give the smallest error. In certain cases, different tips are used for auto zero and the

measurement, this will result in a slightly larger error. In general, the CuveTip is replacing the classical quartz or plastic cuvette and the same principles are applied.

Auto zero and sample measurement with the same CuveTip®

This is the recommended method. The smallest error is achieved when the same CuveTip® is used for auto zero and the sample measurement. This is possible in all cases where cross-contamination between the blank and the sample is not an issue and when cross-contamination between different samples is not critical. For example, in environmental measurements, the auto zero is performed in DI water and measurements are performed directly in e.g. lake water. Another example is the establishment of a calibration curve where the calibration samples are measured from low concentrations to large concentrations using the same tip. If sterile water/buffer is used as a blank this method can be used in cell culture work as well. There are some considerations depending on the sample volume:

If the sample volume is small, below 1 mL, the user must make sure that no blank or sample remains in the CuveTip® cavity. Residues can be removed by cleaning the CuveTip® cavity with a *clean-wipe*. By touching the CuveTip cavity with the wipe, the capillary action of the wipe is used to remove any blank or sample volume from the CuveTip. In this way only a minuscule volume of the old sample is transferred into the new sample.

If the sample volume is large, above 1 mL, the cleaning procedure explained above is not necessary. The small volume transferred into the new sample is negligible compared to the larger sample volume. After inserting the CuveTip into the new sample, the CuveTip must be moved in the sample for ~5 seconds to replace any residues in the CuveTip with the new sample. This will give accurate readings of the sample absorbance.

Auto zero and sample measurement with different CuveTips®

If sample contamination is critical, a new CuveTip® has to be used after auto zero and for any consecutive measurement.

This is usually the case for cell culture and microbial work. Due to the unique design of the CuveTip®, the sample never comes in contact with the Photopette® device. Therefore, contamination and cross-contamination between samples can be totally eliminated by using a new CuveTip® for every measurement.

HOW TO USE THE CUVETIP® CORRECTLY?

There are four steps towards proper use of CuveTip®: 1) Attach the tip, 2) immerse the tip into the blank or sample and measure, 3) retract the tip from the sample, and 4) eject the tip from the device. Its use is simple and convenient; however, some recommendations need to be followed.

Attach

Take the Photopette® device, open the tip box and pick up a new CuveTip® from the box. Ensure that the CuveTip® is adjusted and there is no gap between the tip and the device. Close the tip box to avoid contamination.

Immersion and measurement

Hold the Photopette® device at an angle of 20-30 degree with the CuveTip® cavity pointing upwards. Then immerse the tip slowly into the sample. Immerse the tip at least 3 mm and maximal 15 mm. Check that no air bubble was formed. In case an air bubble is formed, gently tap the CuveTip® onto the wall of the sample container. Proceed with your measurement once the bubble is removed.

Retract

Retract the CuveTip® in the same way it was immersed at a 20-30 degree angle and observe if the liquid flows out from the CuveTip® cavity. In case the liquid does not flow out, tap the CuveTip® gently onto the wall of the sample container. Repeat if necessary. Alternatively, please use a 10 µL micropipette to pull-in the sample.

Eject

Insert the CuveTip® into the ejector box opening. Turn the device at least 60 degree to eject the tip from the stem of the device. The device is now ready for the next measurement.

HOW TO PERFORM AUTO ZERO MEASUREMENT?

The auto zero is performed before measuring the sample. It is the same procedure as used in any other spectrophotometer.

Blank sample

The auto zero is performed in a sample that does not contain the analyte to be measured, this is called a “blank”. The error can be further reduced if the same sample matrix is used for the auto zero, e.g. sterile media without cells for a cell count

measurement. Often, the sample matrix is not available. A media close to the sample should be used in such cases, e.g., buffer or DI water can be used as blank for the auto zero.

WHICH CUVETIP SHALL I USE?

Two different CuveTip® are available, the “CuveTip® Standard” and the “CuveTip® Standard Sterile”. Both tips can be used for UV and visible wavelength; the only difference between the tips is that CuveTip® Standard Sterile is sterilized in a process using ethylene oxide (ETO). For most applications, the non-sterile tips are fine. For cell culture work and any work where microbial contamination must be avoided, we recommend the use of sterile tips. Although, CuveTips® are manufactured with high standards and in clean environment, however the current tips cannot be ascertained to be DNase/RNase free. The user is advised to evaluate contamination risk before performing the measurement.

WHAT IS THE MINIMAL SAMPLE VOLUME AND SAMPLE CONSUMPTION?

Contrary to other spectrophotometers, Photopette® needs an immersion depth and not a minimum sample volume. CuveTip® is a substitute for the classical cuvette and has to be immersed into the sample liquid sample for at least 3 mm. Depending on the container holding the sample, different minimal volumes are required as given in Table 1.

Sample container	Minimum volume
96-well plate	~120 µL
500 µL reaction tube	~120 µL
1.5 mL reaction tube	~150 µL
15 mL centrifuge tube	~1 mL
50 mL centrifuge tube	~1 mL
T75 cell culture flask	~1 mL

Table 1: Minimum required sample volumes in different sample containers.

The liquid sample will flow into the cavity of the CuveTip® when immersed at a minimum of 3 mm. The maximum immersion depth is about 15 mm. The CuveTip® cavity is holding about 40 µL of sample. Due to the adhesion of the sample onto the hydrophilic CuveTip® a volume of ~12 µL remains on the tip and is consumed in every measurement.

A TRICK TO REDUCE SAMPLE VOLUME TO 40 MICROLITER

It is possible to reduce the sample volume to ~40 μL by using the following trick. Attach the CuveTip® and perform auto-zero. By using the same tip or after changing the tip, place the Photopette® device in the stand and pipette about ~40 μL sample directly into the cavity of the CuveTip®. Check that no bubble was formed and perform the measurement as usual. Use the pipette to retrieve the sample from the CuveTip® for reuse.

HOW MANY TIMES CAN I REUSE THE CUVETIP?

CuveTip® is a disposable and is recommended to use “one tip per sample”. If sample cross-contamination must be avoided we strictly recommend to use a new CuveTip® after every measurement. The same applies for sterile tasks where the CuveTip® Standard Sterile should be used and changed for each new sample measurement. For serial measurements, e.g. to establish a calibration curve, the same tip can be used.

CAN I USE THE CUVETIP IN SOLVENTS OTHER THAN WATER?

Yes, the CuveTip® can be used in certain organic solvents such as methanol, ethanol and their mixtures with water. The CuveTip also works in plant or mineral oils. The compatibility for solvents is given in the Photopette® user manual. [1]

AT WHAT TEMPERATURES CAN I USE THE CUVETIP?

The CuveTip® is designed to be used at around room temperature; it can be safely used from 10 to 40 °C. The material of the CuveTip® is a thermoplastic which deforms at higher temperatures. We have performed measurements successfully at higher temperature of up to ~70 °C for a measurement lasting less than five seconds. A user shall perform control experiments to ensure error-free measurements for measuring at high-temperature.

AT WHICH ENVIRONMENTAL LIGHT CONDITION CAN I USE THE CUVETIP®?

The Photopette device has an inbuilt environmental light correction to correct for small changes in the environmental light conditions between the time of auto zero and the measurement. To reduce errors, auto zero and the measurement should be taken at the same location with the same environmental light. If possible, reaction tubes should be placed in a dark holder to shield the sample from environmental light. Photopette is not designed to be used in direct sunlight or close to other bright light sources.

CAN I CLEAN THE CUVETIP®?

The CuveTip® is a high precision optical product with polished windows and mirrors. We do not recommend to

clean the CuveTip®. Cleaning may scratch or otherwise damage the optical surfaces of the CuveTip® and make it unusable for error free measurements.

CAN I USE REAGENTS FROM OTHER COMPANIES WITH THE CUVETIP®?

Yes, you can. Reagents from any company can be used with the Photopette and its CuveTip as long the readout is at a wavelength supported by the Photopette device. Most reagents are made for the 1 mL rectangular standard cuvettes. Advantageously the reagent volume needed for Photopette is much smaller; about 200 μL are more than sufficient. Therefore about 5 times more samples can be measured to save on cost.

DISCUSSIONS

ADVANTAGES

If used correctly, the Photopette® provides ultra-fast, accurate and reliable measurements at the lab bench or in the field. Due to its simple workflow and intuitive iOS and Android app, a non-expert user can perform error-free photometric measurement. The disposable CuveTips® are available as non-sterile and sterile tips to eliminate any chemical and microbial cross contamination of samples.

LIMITATIONS

A minimal sample volume of ~120 μL is required for measurements. With a simple trick highlighted in this application note, the volume can be reduced to 40 μL . A measurement consumes about 12 microliter sample due to its adhesion on the hydrophilic CuveTip®. Unless specified, the Cuvetips® should not be considered as Nuclease-free.

SUMMARY

This application note outlines several tips to make working of Photopette® and CuveTips® easy and convenient. More application specific Application Notes are available on the company's webpage. [3]

REFERENCES

- [1] “Photopette® User Manual V1.0.0” Tip Biosystems Pte Ltd, Singapore, 2016.
- [2] Quick Start Guide, Tip Biosystems webpage at URL www.tipbiosystems.com

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