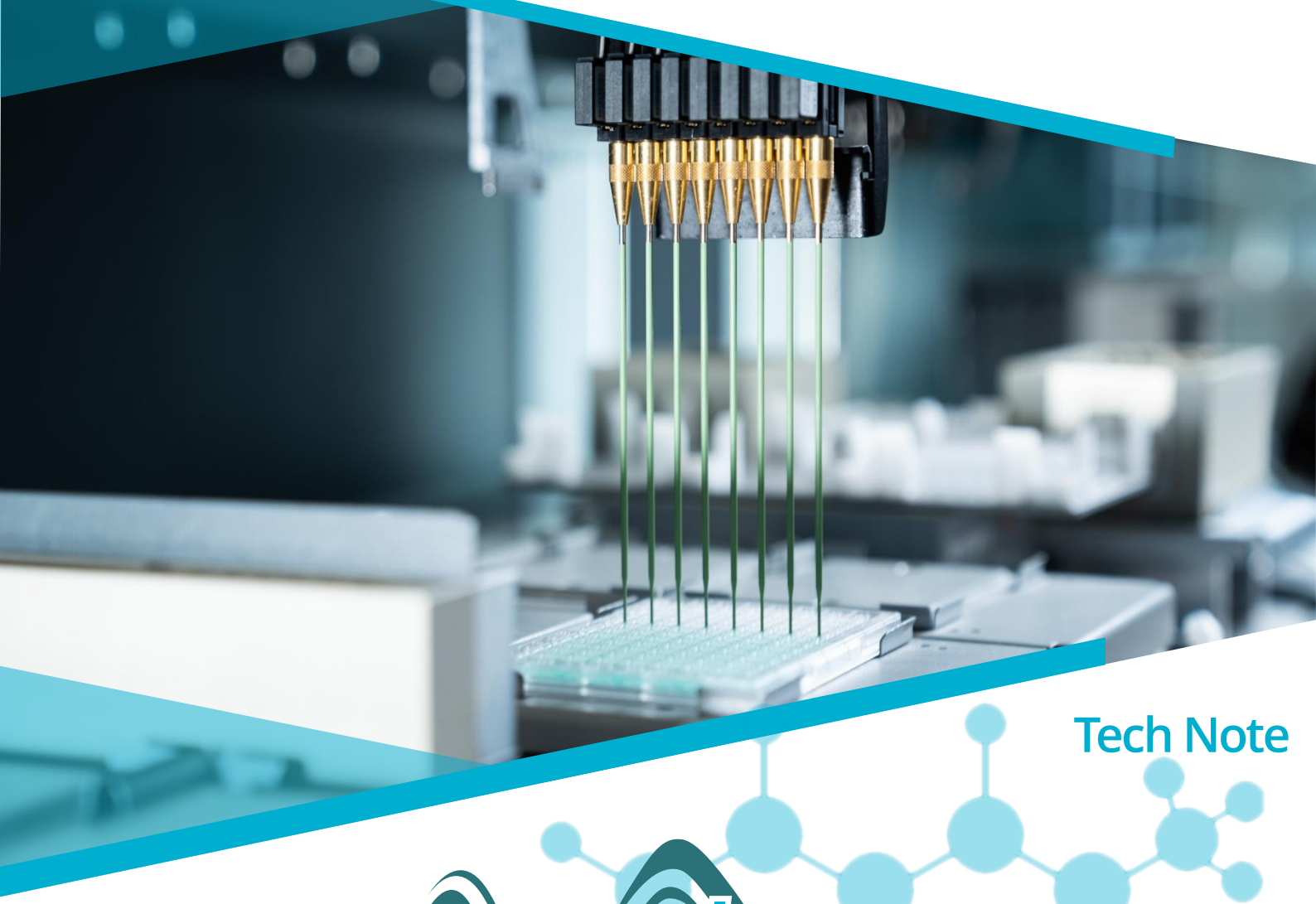

Tech Note

SpecPlate

Best Practices for Manual and Automated Pipetting with SpecPlate

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This TechNote provides important tips and tricks for seamlessly integrating SpecPlate into your existing workflows, whether you use manual or automated filling processes.

Introduction

The SpecPlate has been designed in accordance with the standards set forth by ANSI SLAS, ensuring full compatibility with standard laboratory equipment and seamless integration into existing workflows. The inlets of the SpecPlate structures are positioned in precise alignment with the centers of the wells of a 96-well plate. This allows the use of multichannel pipettes and any liquid handling system designed to handle standard plates. However, filling the SpecPlate may be unfamiliar at first. To facilitate an introduction to the SpecPlate, this Tech Note provides some guidance for manual pipetting as well as filling parameters for automated filling. These parameters can be used as a starting point for implementation in one's liquid handling system.

Material

- SpecPlates (PHABIOC 400100)
- Manual Pipette (according to availability)
- Liquid Handler (according to availability)
- Suitable pipette tips

General Methods

A suitable pipette tip should be used for optimal pipetting pipetting results. For the 36 µL filling volume of a SpecPlate measuring structure, pipette tips within the range up to 300 µL are suitable for manual pipetting. In the case of liquid handlers, the employment of 50 µL tips has proven successful.

It is essential to adjust the flow rate to the specific sample to guarantee uniform filling of the measurement structures. The flow rates recommended in this Tech Note have been established with aqueous solutions and should be used as a preliminary basis for further optimization.

To ensure the complete and accurate filling of all four measuring chambers of the measuring structures, it is imperative that the second pressure point is not pressed when dispensing the sample during manual pipetting (i.e., no blowout) or, in the case of automated pipetting, that no air gaps that may have been taken up prior to aspirating the liquid are dispensed in the measuring structures. Exceptions to this are particular filling patterns (see corresponding section).

The optimal filling results were obtained when the pipette tip was positioned slightly below the waist of the filling opening to dispense the sample (see Figure 1). This waist is situated 2 mm below the upper edge of the filling opening.

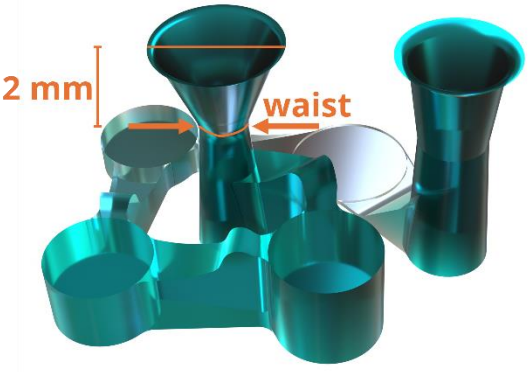


Figure 1 Location of waist in filling opening

Manual Pipetting

Insert the tip of the pipette carefully into the inlet and then gradually and gently release the sample into the structure.

The pipette tip should be inserted vertically or at a slight angle. When inserted at a slight angle, the pipette tip can be supported on the edge of the inlet. It has been shown that when using a multi-channel pipette, vertical insertion is the optimal method.

Automated pipetting

In order to achieve optimal automated pipetting with liquid handlers, it is first necessary to verify the existing plate geometries. As the SpecPlate was developed in accordance with the ANSISLAS standard, it can be assumed that the existing geometries for standard 96-well plates are compatible with the SpecPlate. However, it is recommended to confirm the geometry and make any necessary adjustments.

The parameters listed in Table 1 were determined for successful and reproducible filling with a filling volume of 36 µL using a Tecan Evo with steel tips on an 8-pipette liquid handling arm and with 50 µL disposable tips on a 96-pipette multichannel arm. It should be noted that these parameters serve as a guide and may need to be adjusted from device to device.

Table 1 Filling parameters for SpecPlate on Tecan Evo

Handling Arm	Flowrate
LiHa	30 - 50 µL/s
MCA	20 - 40 µL/s

Particular filling patterns

In certain applications, it may be necessary to fill only the two lower chambers (100 and 700 µm) or only the two higher chambers (1400 and 2000 µm). This reduces the sample volume required to achieve the desired measurement results, thereby increasing the efficiency of the process.

Manual Pipetting: To fill the lower chambers, an initial sample volume of 10 µl is initially pipetted into the inlet. Subsequently, 22-24 µl of air is carefully pipetted into the inlet to achieve optimal sample placement in the lower chambers. A sample volume of 25 µl pipetted into the inlet is sufficient to fill the higher chambers only.

Automated pipetting: When filling with an LiHa of an automated liquid handling station, the necessary volume can be reduced to 25 µL for solely the initial two measuring chambers (2000 and 1400 µm). Furthermore, the flow rate should be decreased to 30 µL/s to ensure uniform filling. When utilizing the MCA, these measuring chambers may be filled with 25 µL at a flow rate of 20 µL/s and a short delay of 10 ms.

Two methods may be employed to fill the lower chambers with the LiHa. One method is to initially dispense 10-12 µL of the sample into the inlet at a flow rate of 30 µL/s. Subsequently, the sample is positioned in the measuring chambers by dispensing air at a volume of 20-25 µL at the same flow rate. If the liquid handling system allows, an air gap (25-27 µL) can be aspirated into the pipette prior to the sample (10-12 µL) being aspirated. This air gap should then be dispensed

following the sample being dispensed. Once the sample and air gap have been dispensed into the measuring structure, a brief delay (2s) and a slow retraction speed of the pipette tips should be implemented. Additionally, the lower measuring chambers may be filled with the MCA through the utilization of an air gap. In this instance, 10-12 µL of the sample is initially dispensed into the measuring structure via the inlet, followed by a 20-25 µL air gap with a flow rate of 20 µL/s and a delay of 2 s.

Conclusion

The methodologies outlined herein facilitate optimal and reproducible filling and utilization of the SpecPlate.

Should you wish to obtain data regarding the parameters of liquid handlers produced by other manufacturers, we kindly request that you contact us.

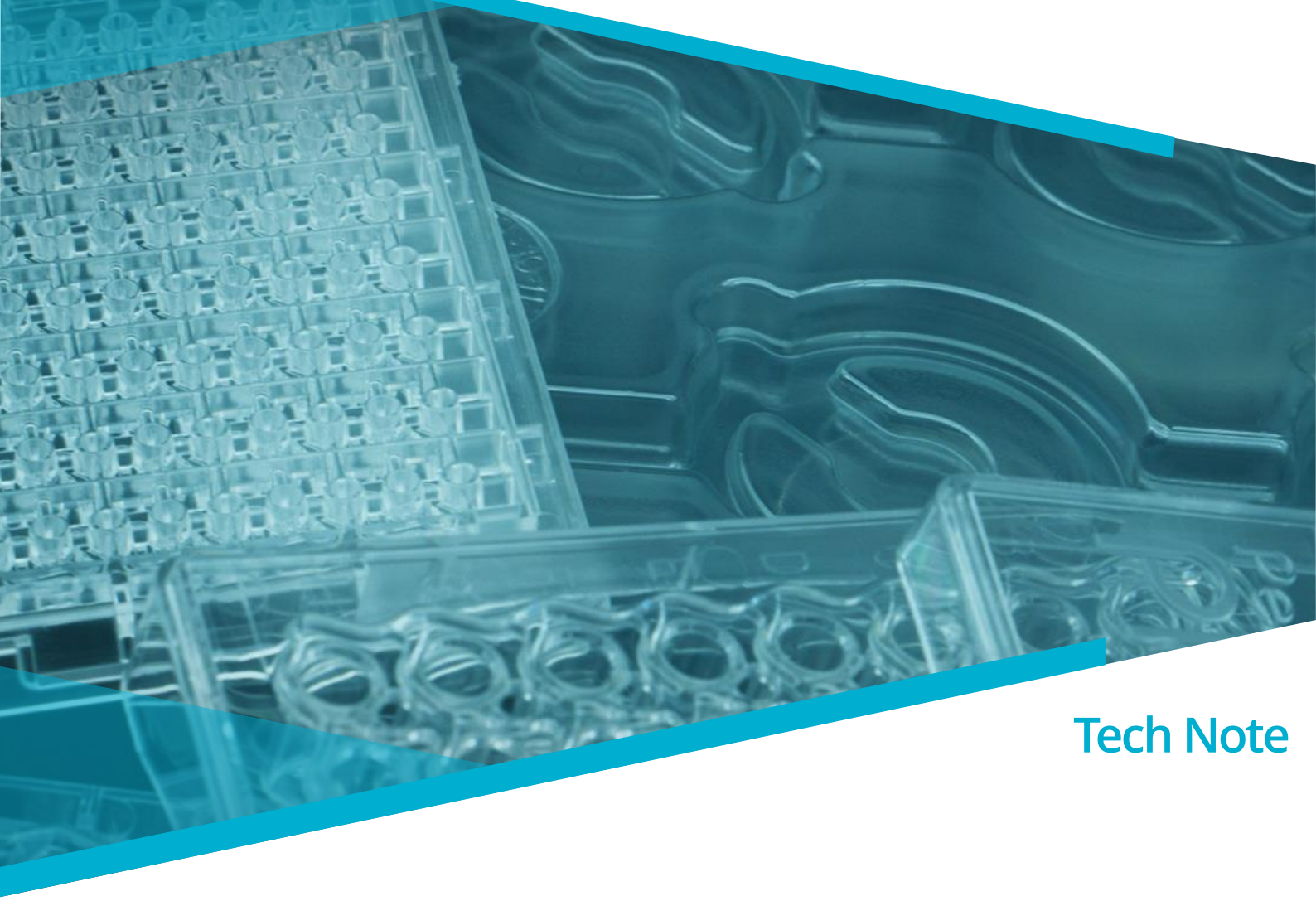
Disclaimer: *It should be noted that PHABIOC cannot be held liable for damage to laboratory equipment resulting from incorrect operation.*

It should be noted that the values provided are mere guidelines and must be tested individually and adjusted if necessary.

The use of SpecPlates is intended exclusively for research purposes.

The results of this study were produced with SpecPlate pre-series models and will be updated with the series products.

Version 1: *Changes, including technical, reserved. 01.09.2024*



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