Reducing the residual volume
while working with the BRAND reagent reservoir 701460
in combination with a 50 µl multichannel liquid end

Abstract
Costly media, solutions, and samples are frequently used in everyday laboratory operations. In order to reduce costs, and minimize the environmental impact, it is of particular interest to work in the most resource-efficient way possible. The BRAND reagent reservoir 701460 is designed to minimize residual volume left behind during pipetting operations, especially for automated pipetting. As a result, it is ideally suited for automation. This technical note describes how the residual volume can be reduced even further while using the BRAND Liquid Handling Station.

Introduction
With the BRAND Liquid Handling Station (LHS), a wide variety of everyday laboratory pipetting tasks can be performed automatically, with accurate results. To prevent the occurrence of inaccuracies and errors, the LHS maintains a safety distance between the pipette tip and the bottom of the vessel, and between the pipette tip and the liquid surface. This ensures that no air is aspirated and no incorrect volumes are pipetted. Both safety distances result in a so-called “residual volume”, which remains in the vessel. Using the BRAND reagent reservoir with cover (Cat. No. 701460) and the 50 µl multichannel liquid end (50 µl MC LE) as an example, this technical note will demonstrate how the residual volume can be reduced through a simple change of the default settings in the LHS software.
Material and method

To reduce the residual volume of the reagent reservoir with the LHS software, the reservoir is first added to the work table. By clicking on the labware, the options available for the labware appear on the right. The residual volume can be reduced in the "Details" tab after "use Custom Tolerances" has been activated (see Figure 1).

The safety distances mentioned previously are the so-called minimum bottom tolerance $X_b$ and the minimum immersion depth $X_i$. $X_b$, indicates the minimum distance between the pipette tip and the bottom of the vessel. The distance will not fall below this value. $X_i$ indicates the minimum immersion depth of the pipette tip into the liquid. The sum of both safety distances is the liquid level that remains when the reservoir is emptied. This liquid level results in a residual volume $V_r$ (Figure 2). $X_b$ and $X_i$ are each preset to 1 mm by default, resulting in a liquid level of 2 mm and, in the case of the reagent reservoir, a residual volume $V_r$ of 666.67 µl with a total volume of 49 ml. To reduce $V_r$, the values $X_b$ and $X_i$ are alternately and successively reduced by 0.1 mm in the software. In the case of selected reservoir, this corresponds to a reduction of the residual volume $V_r$ by 33.33 µl each time. Then, the 50 µl MC LE carries out a pipetting operation to remove the liquid up to the residual volume and to check whether a deterioration of the transfer can be detected. A deterioration manifests itself, for example, when air is aspirated or the pipette tip touches the bottom of the vessel.

In the transfer settings, the LHS is set to dispense above the reservoir so that the tips are more visible (Figure 3).

In addition, prewetting of the pipette tips is deactivated (Figure 4), in order to prevent errors (see Results and discussion). Fully-deionized water (DI water) is used as the medium and the liquid type "Standard" is selected.
Results and discussion

As described above, the values for $X_b$ and $X_i$ were each reduced by 0.1 mm in succession, and as a result the residual volume $V_r$ was reduced by 33.33 µl each time. Following this, a transfer removed the liquid up to the new residual volume and the pipetting quality was tested. The data is presented in the graph in Figure 5.

Right at the beginning, it was apparent that the volume fill level on the left and right of the reservoir is higher than in the middle. This is probably due to adhesion forces with the vessel wall and depends on the liquid in use. It resulted in air being dispensed into the liquid because the tips were prewetted. The subsequent step of aspiration then drew in air along with the liquid. As a result, prewetting the tips was omitted for the entire experiment (Figure 4, right).

The pipetting operation was problem-free up to a residual volume $V_r$ of 400.00 µl (liquid level = 1.2 mm) (see Figure 5). At a considerably lower residual volume, there was a risk of the tips touching the bottom of the reservoir. The potential contact between the tips and the bottom of the vessel is caused by the different tolerances of the liquid end, the reservoir and by inserting the tips, and it illustrates the need for the safety distance $X_b$. As already mentioned, a liquid level of 1.2 mm in this case means that a minimum distance to the bottom $X_b$ of 0.6 mm and a minimum immersion depth $X_i$ of 0.6 mm were selected (compare with Figure 2, right).

From a residual volume $V_r$ of 66.67 µl or less (liquid level ≤ 0.2 mm), the volume was no longer pipetted correctly. Aspiration of air could be observed. This demonstrates that the residual volume $V_r$ can be reduced to 100 µl (liquid level = 0.3 mm) without problems.

In general, however, when reducing the liquid level to between 0.3 and 1.1 mm (residual volume $V_r = 100.00 – 366.67$ µl), the dispensed volume should be checked.

Conclusion

Using the 4 x 40 ml reservoir 701460, the experiment demonstrated how the residual volume $V_r$ is reduced by lowering the minimal immersion depth $X_i$ and the minimum bottom tolerance $X_b$. As a result, the residual volume $V_r$ for the 50 µl MC LE could be reduced from 666.67 µl to 400.00 µl or, as the case may be, to 100.00 µl, depending on the accuracy required. This corresponds to a reduction of the default residual volume setting of approximately 40% (400.00 µl) or, as the case may be, approximately 85% (100.00 µl).
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