Pipette Tip Washing and Other Sustainability Efforts at the National Center for Advancing Translational Sciences (NCATS)

High-throughput robotic platforms help researchers run screening experiments quickly and efficiently, generating results in one week that would take a scientist a decade to conduct manually. At NCATS, these robotic platforms test thousands of potential drugs, drug combinations, environmental chemicals, experimental compounds and biological samples to gain insights about a wide range of human diseases.

For functional genomics screening experiments, 384-well assay-ready plates are routinely prepared with a small volume of siRNA or sgRNA from various library collections. Each of the 384 wells requires a clean pipette tip to enable the transfer of samples from a source library plate to the destination assay-ready plates. Typically, NCATS generates around 2000 assay-ready plates per year, requiring the usage of 768,000 individual pipette tips. This consumable-driven operation was generating a huge amount of plastic trash classified as hazardous chemical waste. This was the driving force for purchasing an automated pipette tip washer, which has resulted in a savings of approximately $240,000 over the last 3 years. Additionally, utilizing washed pipette tips has allowed NCATS to reduce its negative impact on the Earth by reducing waste generation, while producing data indistinguishable from experiments using new pipette tips.

The Grenova TipNovusMini has the ability to deliver cleaning solutions with several customizable options, including sonication, UV sterilization and drying. NCATS has validated their cleaning protocol in two different ways (see poster below) to ensure all data would be comparable with washed pipette tips versus new tips. They have also verified there is no contamination or carryover in the cleaned tips. The functional genomics samples are sensitive and can be easily degraded with contamination due to remnants in the tips, yet no contamination has been observed. An optimized cleaning protocol developed by NCATS has been used successfully on the pipette tip washer for 3 years, allowing for multiple cleanings and re-use of pipette tips, while preserving data quality. The automation team at NCATS took advantage of an opportunity to increase the capability of an existing robotic platform used to cherry pick samples for follow-up experiments by integrating their TipNovusMini. This allows for a fully automated process for pipette tip cleaning for re-use. That said, the same goal of sustainability can be achieved without automation.

In addition to pipette tip washing, the automation team at NCATS has been developing tools, modifying processes and integrating devices to improve their green practices over the last decade. This is particularly important in the high-throughput screening field, which has traditionally relied heavily on consumable plastic labware. For example, a through-beam sensor-based droplet detection system provides notification of clogs or leaks during the automated dispensing protocol, which minimizes the need to re-run experiments. Many biochemical-based screens have been run over the years using a wash and re-use process in place of thousands of assay plates per screen. This has resulted in a savings of $500,000 over several years. NCATS has also developed methods to wash 1536-well assay plates used in cell-based experiments with a device that cleans by way of plasma technology. Their efforts in high-throughput screening waste reduction are always improving and it is their hope that they inspire others to improve their methods as well!

Reduction of Plastic Waste Through the Use of Automated Pipette Tip Washing

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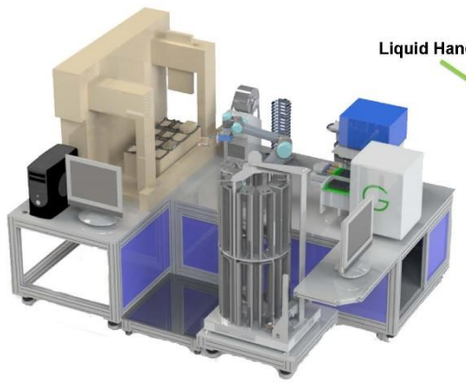
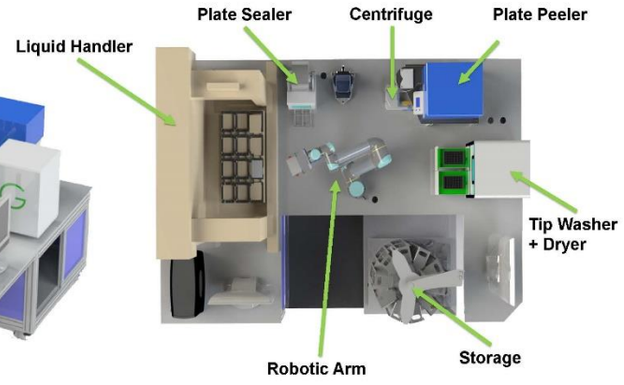
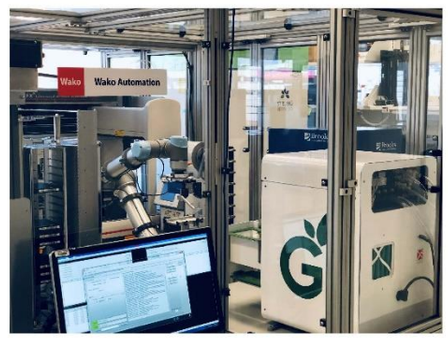
Abstract

Operations of scientific laboratories are largely built upon the accepted use of disposable products. Every day, massive amounts of pipette tips, microplates, cell flasks, and much more are consumed and thrown away after a single use only to be incinerated or tossed into a landfill, in turn, negatively impacting our environment. Ideally, we would minimize this detrimental effect by reducing the amount of waste generated from the assays conducted in our labs while maintaining the integrity of the data produced. Over the last 7 years, the National Center for Advancing Translational Sciences (NCATS) has been working towards minimizing waste from our high throughput screening systems through in-house process adjustments, as well as by evaluating and integrating eco-friendly peripheral devices into our various screening platforms. An example is a pipette tip washer with the capability of cleaning and drying a large variety of pipette tip sizes and brands found in most lab settings. To this end, we have integrated the Grenova TipNovus Mini onto our Wako Automation cherry picking robotic platform in order to automate 384-pipette-tip washing which has helped us to dramatically reduce the waste being generated during siRNA screening library preparation and assay-ready plate stamping in an efficient and cost-effective matter. Comparing siRNA screening results from assay-ready plates prepared using new *versus* washed pipette tips containing the same samples and run in the same assay demonstrates that the data are unaffected by the use of washed pipette tips. Incorporating pipette tip washing into the workflow allows us to maximize the runtime of this robotic system and increases its functionality, thus allowing us to adopt the same methodology of tip washing to other processes. Integration of this pipette tip washer has helped us to take a significant step towards operating in a more environmentally conscious manner while continuing to produce reliable high-quality data.

Wako Automation Cherry Pick System







Integration Solutions

- User will select between landscape and portrait handling depending on the desired labware and process
- The UR5 gripper fingers were modified to allow pick and place of the tip racks into the TipNovus Mini wash and dry drawers. Fingers were angled down to account for the height of the drawer face plate and extenders were added to fit into the 4mm space between either side of the tip box and the adapter
- Shelf was modified to allow the XPeel to sit closer to the UR5 arm since the position was outside of the robot work envelope with the new gripper finger design
- Liconic plate shovel modified to be wider in order to handle the nested styled tip boxes more reliably with the added stability
- Wash and dry drawers treated as individual devices to allow continuous washing and drying for a higher throughput

Future improvements planned

- Proximity sensors in conjunction with Beckhoff I/O will be added to accurately determine if the TipNovus Mini drawers are fully open to avoid crashes
- TipNovus Mini adapters will be redesigned using a material more amenable to automation

Validation Methods

1. 2 uL siRNAs transferred from source 384 library plates to destination assay plates
2. Assay-ready plates stored at -80 °C until day of use then thawed and centrifuged
3. 2 uL negative and positive siRNA controls added into column 23 and 24 respectively (excluding "stripe" experiment)
4. 20 uL of serum free media including 0.03 uL RNAiMAX transfection reagent dispensed into each well
5. Incubation for 30 minutes at room temperature
6. 20 uL cells in 20% serum media dispensed at a concentration of 650 cells/well
7. Incubation for 96 hours at 37 °C, 95% humidity and 5% CO_2
8. 20 uL of CellTiter-Glo dispensed into each well
9. Incubation for 30 minutes at room temperature
10. Luminescence data collected on multimode plate reader for viability

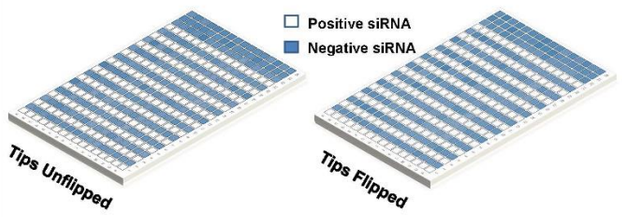
Automated Cleaning Process

1. Tip racks are initially housed in the Liconic labware storage carousel
2. UR5 robotic arm transports the tip rack and places into the wash drawer of the TipNovus Mini; drawer closes and the wash protocol begins (see table below)
3. Once the wash protocol is complete the wash drawer is opened; tip rack is removed and placed into the dry drawer where the tips are dried at 70°C for 12 minutes (simultaneously the next tip rack is moved into the wash drawer)
4. Upon completion of the drying protocol, the tip rack is removed from the drawer and placed back in its original location in the Liconic carousel

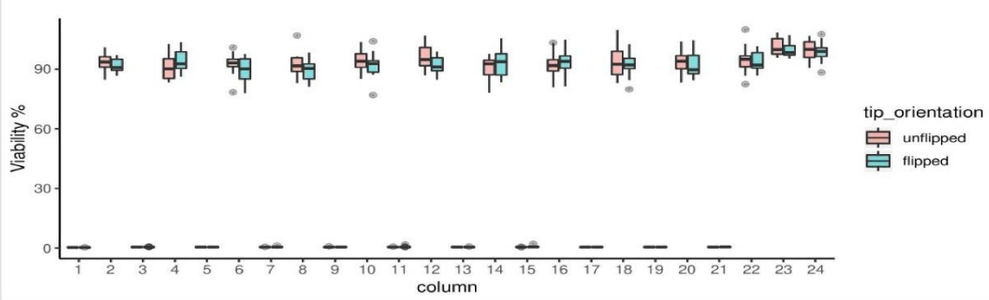
Standard Wash - NCATS

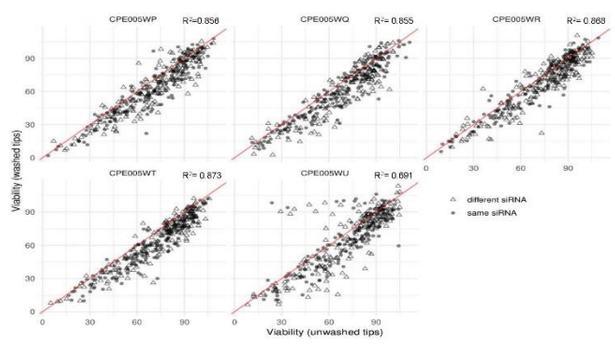
Subprotocol	Reagent	Soak(Y/N)	UV(Y/N)	Sonication (Y/N)	# Purges	Agitation (Y/N)	Volume (L)
Soak Low	DI	Y	Y	Y	4	Y	0.52
Soak High	DI	Y	Y	Y	5	Y	0.65
Pre Wash	Grenoclean	N	Y	N	3	Y	0.39
Wash	DI	N	Y	Y	5	Y	0.65
Rinse	DI	N	Y	Y	3	Y	0.39
Total							2.60

Results



Source plates were prepared containing alternating negative and positive siRNA controls with columns 23 and 24 left empty. Tips cleaned with the TipNovus Mini were used to transfer from the "striped" source plate to the first assay plate after which the tips were washed again. These re-washed tips were used to transfer from the same "striped" source plate but with the orientation reversed. Validation protocol was then followed.





Two sets of assay plates were screened against the Ambion Silencer Select Human Kinase siRNA library to validate the effectiveness of the tip washer. The first set was processed with new sterile tips while the second set was processed using tips cleaned with the TipNovus Mini. Validation assay protocol was then followed.

Conclusions

There is natural synergy in integrating a tip washing device on a platform that utilizes tips to perform cherry pick operations. There is also an improvement in overall system utilization now that the platform can be used both for cherry picking and tip washing operations for a variety of tip type from multichannel pipette systems throughout the center. Data generated using washed tips from the pipette tip washer is of the same quality as data generated using fresh sterile tips directly from the manufacturer. Integration of the tip washer allows for the reduction of material waste while still producing reliable high-quality data and increasing the overall utility of one of our automated platforms.