USER MANUAL

# GlycoWorks™ RapiFluor-MS™ N-Glycan Kits with the Andrew Pipetting Robot

Andrew Alliance × Waters

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## ABOUT THIS MANUAL

This manual describes the installation and use of the Andrew Alliance Andrew Robotic system with the Waters GlycoWorks™ RapiFluor-MS™ N-Glycan Kits. It is an extension of the User Manual of Andrew 1000G-XL and Andrew 1000R-XL.

- ✓ Please read this manual before using the device to ensure proper use and safety.
- ✓ Descriptions are based on the device's default settings.
- ✓ Images and screenshots may differ in appearance from the actual product.
- ✓ Content may differ from the final product, or from software provided by service providers or carriers, and is subject to change without prior notice.
- ✓ Andrew Alliance is not liable for performance issues or incompatibilities caused by misuse of this device.

## INSTRUCTION ICONS

CAUTION

Situation that could cause damage to your device or other equipment

NOTE

Notes, usage tips, or additional information

## TRADEMARKS

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**Patent pending**

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# 1 • INTRODUCTION

This document provides detailed instructions with tips and tricks regarding the general care and use of the Andrew Alliance Andrew Pipetting Robot to perform the manual pipetting actions in conjunction with GlycoWorks™ RapiFluor-MS™ N-Glycan Kits. Andrew is a liquid handling robot that uses conventional pipettes, providing a highly accurate and convenient alternative to manual pipetting and complex liquid handling workstations. The Andrew is comprised of hardware, working deck (called Dominos™), and the Andrew Lab software, intended to make pipetting more reliable and productive.

The Andrew Alliance GlycoWorks™ Package marries semiautomatic glycoprotein sample preparation with fast enzymatic release and rapid labeling of N-glycans.

The GlycoWorks™ RapiFluor-MS™ N-Glycan Kits, enabling unprecedented fluorescent and mass spectrometric performance for glycan detection, are available in 24 and 96 Sample Kits. Depending on sample throughput in the lab, the consumables in these kits can be used for 8-24 samples at a time. Sample number customization is simple with the intuitive Andrew Lab software, if sample needs are variable.

Features of the Andrew Alliance for GlycoWorks RapiFluor-MS offering include the following customized Dominos™/Devices:

- Heating/Cooling Peltier Device [200.799/1, 200.938/2], specially designed to host the 1.2mL reaction tubes that comes with each kit to ensure proper heat transfer.
- Custom GlycoWorks™ RapiFluor-MS™ Reagent Domino™ [200.973/3]
- Waters Vacuum manifold Domino™ [200.978/1], designed to host the Waters 96-Well Plate Extraction Manifold PN 186001831-Refer to Section C.
- Power switch module for automatically controlling SPE vacuum pump [205.114/1]
- Domino™ for Rack of 600µL tubes(Sample Collection Tray) [200.776/1]

# 2 • REQUIRED MATERIAL

## A • TIPS

**Andrew 1000G-XL:**

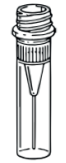
| DESCRIPTION | SUPPLIER REFERENCE | COMMENT | IMAGE |
|---|---|---|---|
| 200µL Extended length tips UltraFine™, FlexTop™. | VWR USA: 37001-522<br>VWR EU: 732-0501 | Extended length tips needed to pipette to the bottom of the 1.2mL reaction tubes. |  |
| 1250µL Extended length tips UltraFine™, FlexTop™. | VWR USA: 53508-918<br>VWR EU: 613-0273 | Extended length 1 mL tips needed for pipetting into the 1.2 mL reaction tubes. |  |



**Andrew 1000G-XL:**

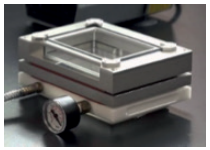
| DESCRIPTION | SUPPLIER REFERENCE | COMMENT | IMAGE |
|---|---|---|---|
| Rainin LTS 250 µL Ext-len. pre-sterilized RT-L250XS tips. | Rainin 17008815 | Extended length tips needed to pipette to the bottom of the 1.2 mL reaction tubes. |  |
| Rainin LTS 1 mL Ext-len. pre-sterilized RT-L1000XS tips. | Rainin 17008818 | Extended length 1 mL tips needed for pipetting into the 1.2 mL reaction tubes. |  |

## B • CONSUMABLES

| DESCRIPTION | SUPPLIER REFERENCE | COMMENT | IMAGE |
|---|---|---|---|
| 0.5 mL screw cap tube with removable cap. | Starstedt #72.785 | These tubes are put into the GlycoWorks *Rapi*Fluor-MS Reagent Domino to hold the solubilized enzyme, *Rapi*Gest and *Rapi*Fluor-MS label. |  |
| Micrew screw cap with X-septum. | Simport PN T347AQX | For 0.5 mL screw cap tube |  |
| 15 mL Conical Centrifuge tube. | Corning #352097 | These tubes are used to hold the reagents for the SPE steps. |  |

| DESCRIPTION | SUPPLIER REFERENCE | COMMENT | IMAGE |
|---|---|---|---|
| 50 mL Conical Centrifuge tube. | Corning #352070 | These tubes are used to hold the reagents for the SPE steps. |  |
| 96 PCR plate without skirt. | VWR 72.1978.202 | For source protein sample |  |

## C • ADDITIONAL MATERIAL

| DESCRIPTION | SUPPLIER REFERENCE | COMMENT | IMAGE |
|---|---|---|---|
| 96-well Plate Extraction Vacuum Manifold | Waters 186001831 | The vacuum manifold is used to purify the sample via µElution SPE. |  |
| SPE Vacuum Pump | 220V/240V 50Hz Waters 725000604<br><br>110/115 V 60H Waters 725000417 | Used with the vacuum manifold. |  |
| Vacuum Manifold Shims (set of 3) | Waters 186007986 | The SPE steps of this protocol will require the use of shims for optimal positioning of components in a vacuum manifold. |  |



| DESCRIPTION | SUPPLIER REFERENCE | COMMENT | IMAGE |
|---|---|---|---|
| USB HUB | Trendnet TU3-H4E | Depending on the number of available USB ports your computer has, it might be required to use a USB port extender in order to plug in Andrew, the INHECO command unit (heating/cooling Peltier device) and Vacuum Power switch.<br><br>A USB HUB is included in Andrew PRO packages. |  |
| Multiplug | For US: Tripp-Lite ISOBAR4ULTRA<br><br>For EU: Any | Suggested since PC, Peltier controller and Andrew must be powered on |  |

## D • SOLUTIONS

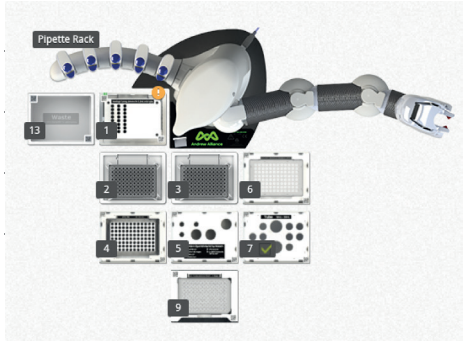
| DESCRIPTION | RECOMMENDED SUPPLIERS |
|---|---|
| Formic acid | LC-MS-grade formic acid is recommended. Fisher p/n A117 or equivalent. |
| 18.2 MΩ water | For glycan sample prep. LC-MS-grade water is critical for glycan analysis. Fisher p/n W6 or equivalent. |
| Acetonitrile | LC-MS-grade acetonitrile is highly recommended for mass spectrometry analysis of glycans. Fisher p/n A955 or equivalent. |
| Ammonium Formate Solution | Recommended use is Waters (p/n 186007081), or use Fisher p/n A115. |

# 3 • IQ/OQ

## A • INTRODUCTION

**Protocol name :**
"IQ_*OQ for Glycoworks kit_Andrew model.anp*"
After installation of your Andrew unit, it is important to perform the IQ/OQ process in order to make sure the system performs as expected. In more detail, the IQ/OQ protocol allows you to check the:

- Correct manipulation of pipettes (Andrew 1000G-XL: P20, P100, P200, P1000 or Andrew 1000R-XL: L100, L200, L1000)
- Qualitative verification of dead volume in 96 PCR plate and 1.2 mL strip tube
- Alignment in 96 PCR plate and 1.2 mL strip tube
- Control of Peltier Domino™
- Control of pump for Vacuum Domino™




**DECK POSITIONS (LAYOUT: NARROW)**

1 • Peltier Domino™
2 • 1000µL Pipette Tips
3 • 200/250µL Pipette Tips
4 • 96-well PCR Domino™
5 • GlycoWorks™ RapiFluor -MS™ Reagent Domino™
6 • Domino™ for Rack of 600µL tubes (Sample Collection Tray)
7 • 15/50mL Tube Domino™
9 • Vacuum manifold Domino™
13 • Waste

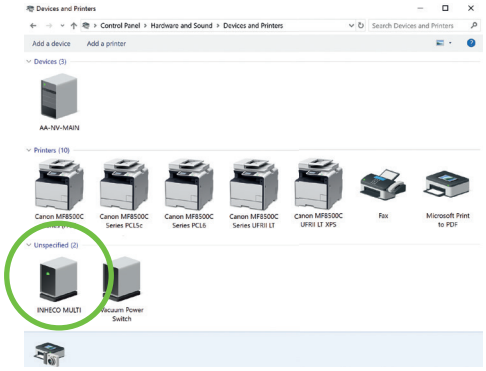
The waste is in position 12 or 13 according to if your Andrew has been calibrated with a Liquid Level Wand (LLW) device or not.

The following Dominos™ are not used in the IQ experiment, but required for the Domino™ allocation:
• GlycoWorks™ RapiFluor -MS™ Reagent Domino™ [200.973/3]
• Domino™ for Rack of 600µL tubes [200.776/1]
• Waters Vacuum manifold Domino™ [200.978/1]

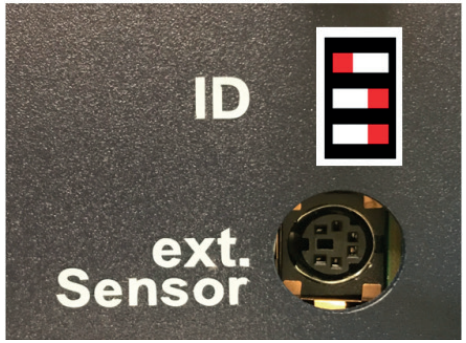
## B • STEPS TO BE CHECKED

✓ Make sure the pipettes placed on Andrew are correctly validated/calibrated.
✓ Connect the Peltier Controller to the power socket by using its power cord.
✓ Connect the Peltier Domino™ with the Peltier Controller by using the green serial cable, as well as the Peltier Controller with the computer, by using the provided USB cable.


✓ Verify the location of the green serial cable from the Peltier controller to the Domino™. It mustn't be on the working area of Andrew.
✓ Verify that the Peltier controller is ON and connected to the PC.



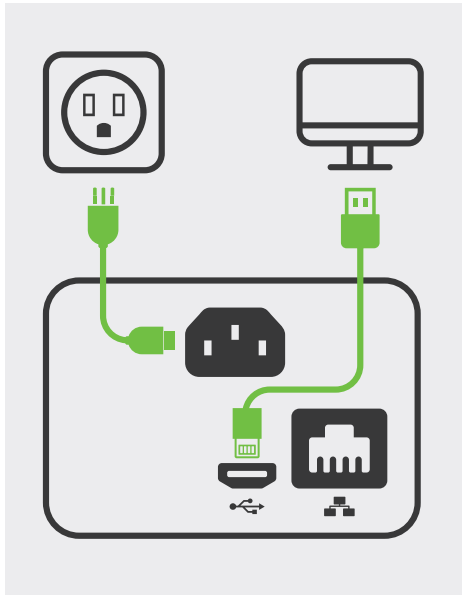

✓ Verify that the ID slot on the back of the Peltier controller is set as follows:

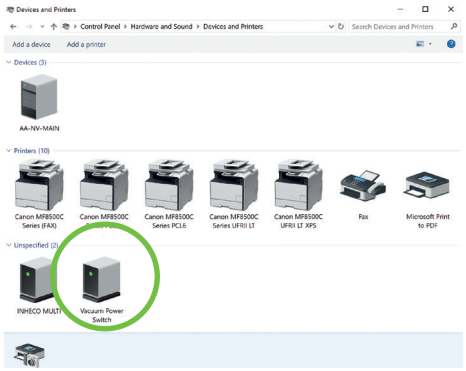



✓ Check the performances of vacuum manifold when the µElution™ plate is present on the Domino™. The vacuum should be between 2.5–4 Hg. If not, you may need to adjust the pump accordingly.

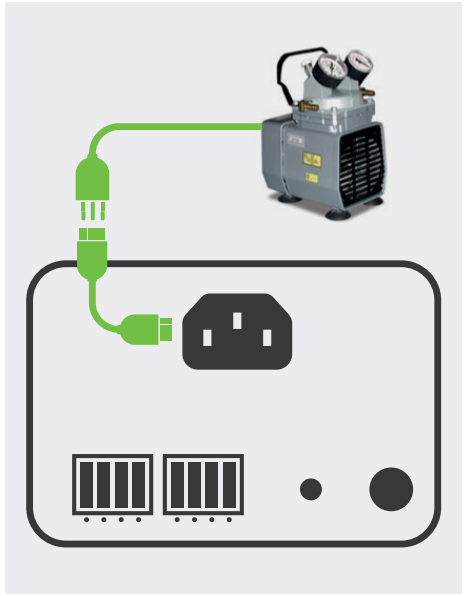
For this step the pump must be powered directly from the wall (not through the power switch).

✓ Verify that the Vacuum Power Switch module is connected to the power and free USB port of your PC/USB Hub.






✓ Unplug the pump from the wall and connect it directly to the A/C Output port of the Vacuum Power Switch by using the provided extension cord as shown below:



✓ Verify that the vacuum pump is placed on the floor or on a different table than Andrew.

When the pump is ON it could generate vibrations, which may influence Andrew during the experiment, if placed on the same workbench.

✓ Verify the absence of tension in the tube that connects the vacuum pump to the vacuum manifold.

✓ Make sure the specially designed shims are placed on the bottom of the vacuum manifold.

✓ At the beginning of the experiment, a waste consumable must be placed inside the vacuum manifold domino, beneath the µElution™ plate.

✓ Run the protocol and validate visually each requested user action.

# 4 • SETTING UP THE ANDREW ROBOT FOR USE WITH THE GLYCOWORKS RAPIFLUOR-MS N-GLYCAN KITS

## A • LOADING THE PROTOCOL/SCRIPT ON ANDREW LAB

Two protocols are included on the computer that comes with the Andrew Alliance GlycoWorks package, one for 8 samples and one for 24 samples. When purchasing reagents for the 8 or 24 sample protocol the user will receive enough reagent to perform the protocol multiple times and as such, these protocols are referred to in this document as "QC 24" (8 samples X 3) and "QC 96" (24 samples X 4). These protocols are similar to the methods found in the GlycoWorks Care and Use manuals (p/n 715004903 for 24 samples; p/n 715004793EN for 96 samples). Some steps have been optimized to minimize total protocol time by maximizing pipetting during incubation steps. Additionally, reagent concentrations and pipetting volumes have been modified from those found in the GlycoWorks Care and Use manuals to match the conditions described in the Waters Application Note *Quality Control and Automation Friendly GlycoWorks RapiFluor-MS N-Glycan Sample Preparation.*
The user should check over the Andrew Lab protocol prior to setting anything up on the deck.

When the pump is ON it could generate vibrations, which may influence Andrew during the experiment, if placed on the same workbench.

Ensure that the sample cleanup and elution steps are performed in the correct µElution™ plate wells and that the dilution steps at the end are in the correct sample collection tubes.

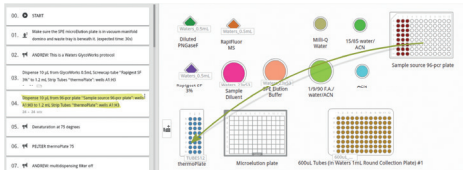
The below steps need to be switched individually in order to comply with the desired source/destination wells:

i. Well prep steps (water & 15/85 water/ACN)
ii. Sample addition to µElution™ plate
iii. 1/9/90 formic/water/ACN x 2 steps
iv. Elution buffer x 3 steps
v. ACN dilution
vi. Sample diluents addition

## B • NORMALIZATION FEATURE (OPTIONAL)

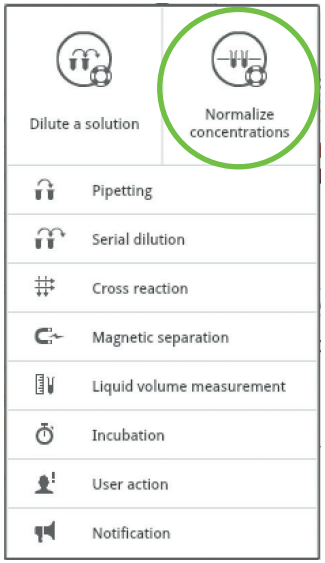
If adding a normalization step to the beginning of the protocol is required, follow the below steps:

**1•** Delete Step 04, as well as *Sample source 96-pcr* plate consumable

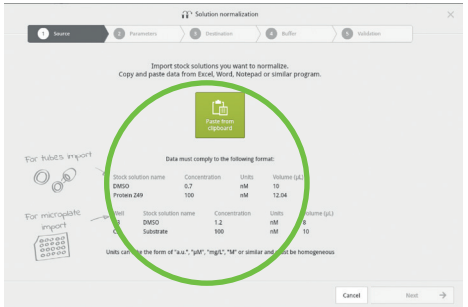


**2•** Click on *"Step 02. ANDREW: This is a Waters GlycoWorks protocol"* step of Andrew Lab's activity log.

**3•** Click on *"Add Step"* button of your Andrew Lab and select the *"Normalize Concentrations"* wizard.




**4•** Paste the sample well, name, concentration etc. following the specific format, which is given to you in the Andrew Lab software.



A .txt or Microsoft Excel file is needed to copy and paste the information into Andrew Lab from your clipboard. Excel may need to be loaded onto the computer being used. If Excel is not available, you may use Google Sheets to copy and paste from.

Include only the data, not the column headers, when copying and pasting the information into Andrew Lab.

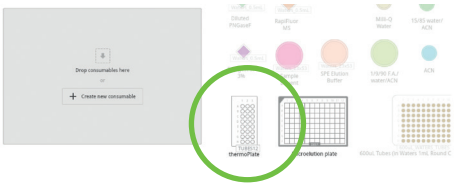
**5•** You will then be prompted to define the desired plate type (ex. normal PCR plate can be found as *96-pcr plate* - 200.176/1)

**6•** Select *"Next"*. Andrew Lab will ask for the desired concentration and volume.

a. For QC the concentration will be 1.5 mg/mL and the volume will be 10 µL.

b. Ensure *"moving an aliquot into another consumable and subsequent dilution"* option is checked.

**7•** On the next page, select the 24-well *thermoPlate-TUBES12* consumable as the destination and into which wells you want sample to be diluted (this depends on the sample number you would like to perform).



**8•** The next page will prompt you to select which buffer you want to use to dilute the samples. On the GlycoWorks Reagent Domino there is a location for the GlycoWorks Rapid Buffer-5ml solution (PN 186008100). Thus, to use this consumable, select from the dropdown consumable list of Andrew Lab the *GlycoWorks Rapid Buffer-Waters_Buffer* consumable. Then, fill the consumable with stock solution (ex. Buffer) and leave the volume field on AUTO.

**9•** Click *"Done"*. This will insert the normalization step as the first step in the protocol.

## C • SETTING UP THE ANDREW

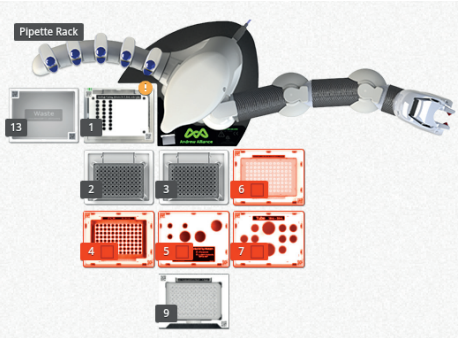
**1•** Once the robot is plugged in and the correct protocol is open, hitting "Execute" will start the protocol.

This step could take a minute for Andrew to initialize its hardware. In case an error appears during this process stating that dust has been detected on the mirror of the Optical Localization Module (OLM), use a Kim-wipe or compressed air to clean the mirror and try again.

Ensure that the Andrew Pipetting Robot placement has bench clearance as its robotic arm will extend and swing over the OLM area.

2• Andrew Lab will show you the Domino™ setup required.

3• Select the Deck layout as *Narrow.*




**DECK POSITIONS (LAYOUT: NARROW)**

1 • Peltier Domino™
2 • 1000µL Pipette Tips
3 • 200/250µL Pipette Tips
4 • 96-well PCR Domino™
5 • GlycoWorks™ RapiFluor -MS™ Reagent Domino™
6 • Domino™ for Rack of 600µL tubes (Sample Collection Tray)
7 • 15/50mL Tube Domino™
9 • Vacuum manifold Domino™
13 • Waste

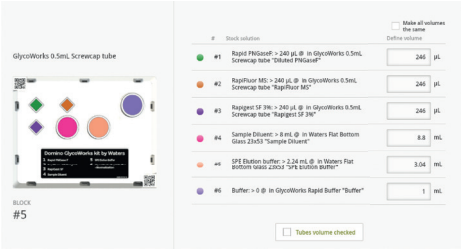
4• Move the Dominos™ to their correct location following the guidelines from Andrew Lab.

Use caution when adjusting the µElution™ Plate Domino™. At the beginning of the experiment a waste tray should be placed beneath the µElution™ plate. Later in the protocol the user will be asked to remove the top of this Domino™ to insert the sample collection tray. The Domino ™ should be returned to the exact location in which it started. To ensure this, it is suggested to align the Domino™ using the two protrusions on the back and note their location to ensure proper alignment throughout the protocol.

5• Selecting each Domino™ position in Andrew Lab will highlight what equipment should be present. Dominos™ that are marked red indicate that the user must define a volume for a solution or check that the volume is correct. You may use the liquid level charts present on the sides of your Dominos™ to check the volume that is present and define it accordingly in the experiment preparation phase.

At the beginning of the experiment the Rack of 600µL Tubes Domino™ in position 6 will indicate that there should be 90 µL of "Labeled N-Glycans" in every well of the plate. This is meant as a placeholder for when the sample collection tray is moved out of the vacuum manifold and into its own Domino later in the protocol. The sample collection tubes (placed in their correct wells) should be left empty, while its stock solution should be virtually confirmed.

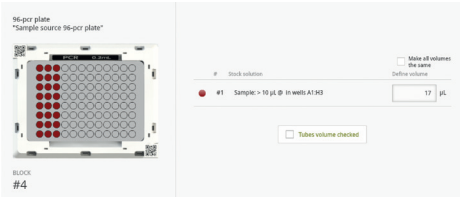
6• Once all solutions in the Dominos™ are accurate, select the check box at the bottom of the solution list that signifies they've been checked.




## D • REAGENT PREPARATION

***Intact mAb Mass Check Standard***

Use of a control standard is highly recommended.
Reconstitute 1 vial (1 mg/vial) of Intact mAb Mass Check Standard using
QC 24/96: 666.7 µL of 18.2 MΩ water to create a 1.5 mg/mL IgG solution.




***RapiGest SF Surfactant***

QC 24: Prepare 3% (w/v) *Rapi*Gest SF by dissolving 3 mg of *Rapi*Gest SF Surfactant in 60 µL of GlycoWorks Rapid Buffer and 40 µL of water. Vortex to mix for 10–15 seconds.

QC 96: Prepare 3% (w/v) *Rapi*Gest SF by dissolving 10 mg of *Rapi*Gest SF Surfactant in 200 µL of GlycoWorks Rapid Buffer and 135 µL water.
Vortex to mix for 10–15 seconds.
Transfer entire volume to a low-dead-volume 0.5 mL tube with the green x-septa caps from Simport.

Pre-slit the cap by keeping the cap ON THE TUBE when transferring sample into the vials and poking through the septa.

***GlycoWorks Rapid PNGaseF Enzyme***

QC 24/96: Dilute PNGase F Enzyme (30 µL) with 220 µL water for a total of 250 µL. Transfer entire volume to a low-dead-volume 0.5 mL tube with the green x-septa caps from Simport.

Pre-slit the cap by keeping the cap ON THE TUBE when transferring sample into the vials and poking through the septa.

Please note that the glycoprotein denaturation and PNGase F digestion protocols provided, were designed to be effective for the released N-glycan analysis of monoclonal antibodies and other glycoproteins with low to moderate N-glycan antennarity and sialylation, as well as disulfide bonding patterns that do not significantly sterically restrict access of the PNGase F enzyme to the N-link site. In the event that this procedure does not provide complete glycan release, it is recommended to consult the GlycoWorks RapiFluor-MS N-Glycan Kit Care and Use manual for method adaptations that can be used to provide accurate results for those glycoproteins.

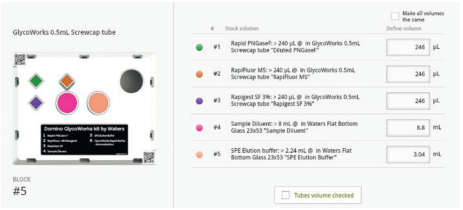
***RapiFluor -MS***

QC 24: Add 110 µL of anhydrous DMF directly to one vial of 9 mg of RapiFluor -MS Reagent. Vortex mix to solubilize.
QC 96: Add 280 µL of anhydrous DMF directly to one vial of 23 mg of RapiFluor -MS Reagent. Vortex mix to solubilize.

Vortexing is a critical mixing step; otherwise some of the RapiFluor -MS powder will remain at the top of the tube.

Transfer entire volume to a low-dead-volume 0.5 mL tube with the green x-septa caps from Simport.

Pre-slit the cap by keeping the cap ON THE TUBE when transferring sample into the vials and poking through the septa.




***GlycoWorks SPE Reagents***

Load the Elution Buffer and Sample Diluent included in the GlycoWorks SPE Module in the correct slots in the Reagent Domino.

The vials are similar enough that they're easy to mix up. Andrew Lab will have them as different colors in the domino.

***SPE clean-up solutions***

Common chemical solvents are not included. Prior to beginning this protocol, the user will need to prepare the following solutions for the SPE clean-up module:

1• 18.2 MΩ water in a 15 mL Falcon tube.
2• Acetonitrile in a 15 mL Falcon tube.
3• 15:85 (v/v) 18.2 MΩ water/acetonitrile in a 15 mL Falcon tube.
4• 1:9:90 (v/v/v) formic acid/18.2 MΩ water/ acetonitrile in a 50 mL Falcon tube.

Volumes should be measured out individually and accurately (±3%) and then combined to make solutions 3 and 4 for the clean-up step.

| DESCRIPTION | | | | 8 SAMPLES | 24 SAMPLES |
|---|---|---|---|---|---|
| Domino | Reagent | Consumable | QC | QC | QC |
| 15 mL and 50mL Falcon tube | ddWater | 15 mL Falcon | ≥2.25 mL | ≥5.25 mL | |
| | ACN | 15 mL Falcon | ≥3.25 mL | ≥9.25 mL | |
| | 15:85, Water:ACN | 15 mL Falcon | ≥2.25 mL | ≥5.25 mL | |
| | 1:9:90, F.A.: Water:ACN | 50 mL Falcon | ≥10.1 mL | ≥29.3 mL | |
| GlycoWorks | PNGase F | 0.5 mL tube w/ septa cap | ≥86 µL | ≥246 µL | |
| | Domino | 0.5 mL tube w/ septa cap | ≥86 µL | ≥246 µL | |
| | RFMS | 0.5 mL tube w/ septa cap | ≥86 µL | ≥246 µL | |
| | Sample Diluent | Waters glass vials | ≥3.8 mL | ≥8.8 mL | |
| Microplate | Elution Buffer | Waters glass vials | ≥1.58 mL | ≥3.04 mL | |
| | Protein samples | 96-well PCR plate | ≥17 µL/well | ≥17 µL/well | |
| Tips | N/A | D200 | ≥27 | ≥75 | |
| | N/A | D1000 | ≥14 | ≥30 | |



## E • RUNNING THE PROTOCOL

1• Check to ensure that all pipettes are set up in the correct slots in the arm.

Vortexing is a critical mixing step; otherwise some of the RapiFluor -MS powder will remain at the top of the tube.

2• Make sure all solutions are checked over and confirmed to be the correct volume (see section 4.C. and 4.D.),

3• Click on each tip box Domino™ icons in Andrew Lab to know how many tips are needed for the amount of samples you are running.

4• Click on "Start" to begin the protocol.

5• At this point Andrew checks that every Domino™ is in the correct location and that there are enough pipette tips to complete the protocol. The tip plugs need to define a COMPLETED rectangle of tips, as shown in Andrew Lab.

Certain tips, like 200 µL tips, can cause a failure during the Domino scan process. The way these tips sit in the box can make it difficult for Andrew to find the code on the plugs. Resolve this failure by making sure the plugs are flat in the holding tips, then start the process again.

6• Once Andrew starts the protocol, the first move is to measure where the end of the pipette is without a tip on it. This is done the first time the Andrew robot picks up each of the 5 pipettes in the set when performing the protocol to achieve a smooth tip insertion. Then, it will measure the properties of each tip to ensure proper alignment and go ahead with each protocol step.

## F • SPE CLEAN-UP OVERVIEW

Three user interactions are written into the protocol at the SPE steps, specifically between the µElution plate steps. This is a precautionary measure.

1• The first user interaction occurs at the beginning of the experiment. The protocol pauses to make sure the user has inserted the waste tray in the µelution™ plate Domino™. This will ensure the system is ready when the steps for conditioning the wells with water, equilibrating the sorbent with 15/85 water/ACN, dispensing sample, and washing sampleon the µelution™ plate need to take place.

2• The second user action requires the user carefully remove the top of the vacuum manifold, replace the waste tray with the sample collection tray loaded with 600ul sample collection tubes, replace the top of the vacuum manifold in the exact orientation it was previously, and remove the cap from the SPE Elution Buffer reagent.

3• The third and final user interaction occurs once the labeled sample is fully eluted. The user is required to move the sample collection tray from the vacuum manifold into the empty Rack of 600µL Tubes Domino™ for the dilution steps.

After each user action, the user is required to press the "Resume" button to continue with the protocol.

## G • SAMPLE ANALYSIS

Once the dilution is finished, the sample tubes can be capped and are ready for analysis.

Aspiration has been found to not thoroughly mix each sample. An obvious sign that this is occurring is non-reproducible N-glycan chromatographic peak areas. **It is recommended that the user vortex their samples for at least 10 seconds to ensure mixing.**

# 5 • STORAGE AND STABILITY OF SOLUTIONS

| STANDARD/REAGENT | INITIAL STORAGE | AFTER RECONSTITUTION | SOURCE |
|---|---|---|---|
| Intact mAb Check Standard | Freeze on arrival (-20 °C) | 10 °C for 1 week or sub-aliquot and freeze. AVOID freeze/thaw cycles. | Intact mAb Mass Check Standard Care and Use Manual(720004420en) |
| GlycoWorks RapiFluor-MS Performance Test Standard (Optional PN) | Freeze on arrival (-20 °C) | 4–10 °C for 1 week or sub-aliquot and freeze (-80) for 3 months. AVOID freeze/thaw cycles. | RapiFluor -MS Performance Test Standard Care and Use Manual (720005349en) |
| GlycoWorks Rapid PNGase F and GlycoWorks Rapid Buffer | Fridge (2–8 °C ) | Rapid PNGase F and GlycoWorks Rapid Buffer should be stored at 2–8 °C for up to 24 months. AVOID freeze/thaw cycles. Bring to room temperature and centrifuge before use. | GlycoWorks RapiFluor -MS Kit Care and Use Manual (715004793en) |
| RapiGest SF | Room temperature | Once reconstituted in high purity water or a buffer (pH 7–10) the solution is stable for one week when stored at 2–8 °C. Long term storage of frozen aliquots is possible but not recommended due to potential solubilization issues of RapiGest SF or storage buffer. Note: RapiGest SF hydrolyzes in acidic solutions (half life 8 min at pH 2 and 60 min at pH 3). | RapiGest Care and Use Manual (715000122en) |
| RapiFluor -MS Label | Room temperature | It is recommended to store the plastic paraffin film vial in a sealed bag along with a desiccant pack at room temperature. Once solubilized, the RapiFluor -MS reagent solution should be stored at -80 °C (the freezing point of DMF is -61°C) under anhydrous conditions. It is suggested to store RapiFluor -MS reagent solution as 12 µL aliquots in 600 µL microcentrifuge tubes and to subject labeling reagent aliquots to only a single freeze-thaw cycle. In order to further minimize contamination of the reagent solution with water condensation, the analyst must allow aliquots to equilibrate to room temperature before the aliquot tube is opened. | GlycoWorks RapiFluor -MS Kit Care and Use Manual (715004793en) |



| STANDARD/REAGENT | INITIAL STORAGE | AFTER RECONSTITUTION | SOURCE |
|---|---|---|---|
| GlycoWorks Reagent Solvent Anhydrous DMF Packaged with Label | Room temperature | Once the ampoule is opened, it needs to be used right away to solubilize the label. | GlycoWorks RapiFluor -MS Kit Care and Use Manual (715004793en) |
| GlycoWorks SPE Reagents (SPE Elution Buffer and Sample Diluent) | Room temperature | Stable until expiration date. If opened many times, it is good to check pH of elution buffer. Once SPE Diluent is opened use it right away to avoid ACN evaporation to minimize chromatography solvent effects. | GlycoWorks RapiFluor -MS Kit Care and Use Manual (715004793en) |
| GlycoWorks HILIC µElution Plate | Room temperature-dry | After partial use store in the open pouch, squeeze out any air, fold over the open end of the pouch and seal with tape. Store in a desiccator. | GlycoWorks RapiFluor -MS Kit Care and Use Manual (715004793en) |
| Ammonium Formate Solution (Mobile Phase A Concentrate) – (Starter Kits or Optional PN) | Room temperature | Room temperature for no longer than one month once solubilized. When not in use store it in the fridge – ensuring time to get to room temp upon before re-use. For smaller volume needs, one could use 1 mL at a time to add to 100 mL volumes of water. | Product COA |
| ACQUITY UPLC Glycan BEH Amide Column | Room temperature | For column regeneration and storage: Instead of using 100% aqueous mobile phase for regeneration, a 75/25 composition can be used as a means to extend the column lifetime. However, it should be kept in mind that some applications may require a 100% aqueous regeneration step in order to minimize sample derived buildup. | Care and Use Manual (720003042EN) |

# 6 • RESOURCES AND REFERENCES

**ANDREW SYSTEM:**
Andrew Lab User Guide
Andrew Robot User Guide

**CARE AND USE MANUALS/QUICK START GUIDES FOR QC PROTOCOL:**

GlycoWorks RapiFluor-MS N-Glycan Kit – 96 Sample Care and Use Manual literature code 715004793EN

GlycoWorks RapiFluor-MS N-Glycan Kit – 24 Sample Care and Use Manual, literature code 715004903

RapiFluor-MS Quick Start Protocol – 96 Sample, literature code 720005343EN

RapiFluor-MS Quick Start Protocol – 24 Sample, literature code 720005470EN

GlycoWorks Ordering Information and Product Guide, literature code 720005708EN

**APPLICATION NOTES:**
Quality Control and Automation Friendly GlycoWorks RapiFluor – MS N-Glycan Sample Preparation literature code 720005506EN

**INFORMATIONAL VIDEO:**
Waters and Andrew Alliance Partnership for Analytical Laboratory Automation:
https://youtu.be/FBWJUFAVwPk


Andrew Alliance