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Protocol status: Working
We use this protocol and it's working

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🌐 Feedstocks-to-Fuels Labman solid biomass dispensing protocol

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DISCLAIMER

ABSTRACT

This protocol details the steps necessary for aliquoting biomass for the Feedstocks-to-Fuels pipeline. Samples must be ground and put in scintillation or Sarstedt tubes prior to dispensing into the Micronic48 tubes. The tolerance is typically below 10%, but check the settings for the dispensing method you are using. Be sure to grind the biomass thoroughly to minimize clogs

IMAGE ATTRIBUTION

Icon graphic created with BioRender.com

GUIDELINES

- This protocol requires dispensing methods that are specific to a particular biomass and a particular distribution amount. Confirm that the correct dispensing method is available prior to running this protocol.

- A 2mm grind or smaller is recommended for all biomass to prevent the robot from clogging.

- If the biomass of interest has a tendency to form needles or a tendancy to "clump", performing a grinding protocol on the Labman prior to running this method is recommended.

- As a general rule of thumb, a full scintillation tube can hold ~4.5g of biomass. To ensure success, do not map more than 4g of biomass from a full scintillation tube to destination tubes. This means, for example, If you are adding ~0.2 g per destination tube, then each source tube should map to no more than 10 destination tubes.

- It takes ~ 1 min to distribute ~0.2 g of biomass into each Micronic48 tube. This means that it will take ~ 1 hour to complete a full plate.

MATERIALS

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⊗	Screw Cap Micro Tube 2 mL polypropylene (PP) **Sarstedt Catalog #72.694.005**
⊗	Kimble™ 20mL Glass Screw-Thread Scintillation Vials **Thermo Fisher Catalog #22043725**

- Ground biomass in 20mL plastic scintillation vials or 2mL Sarsvedt tubes
- 48 tube micronic rack

SAFETY WARNINGS

> ❗ Some ground biomass can become airborne when being distributed, leading to potential inhalation and irritation. Please wear an N-95 mask when dispensing biomass.

BEFORE START INSTRUCTIONS

You will need:

- Dispensing method for your type of biomass
- Input and mapping files
- Ground biomass distributed into scintillation vials or Sarstedt tubes

Software Layout and Start Up

`1m`

`1m`

1 The graphical layout of this user interface (UI) is intended to resemble the deck of the Labman if you were facing the deck on the same side as the monitor.

- If the software is already open when you start your run: click on the "**Clear Racks**" button of the "Function Buttons". This will clear out settings from the previous run

- If you opened the software yourself, click on the "**Initialize**" button of the "Function Buttons". This will allow all electronics to calibrate prior to the run.
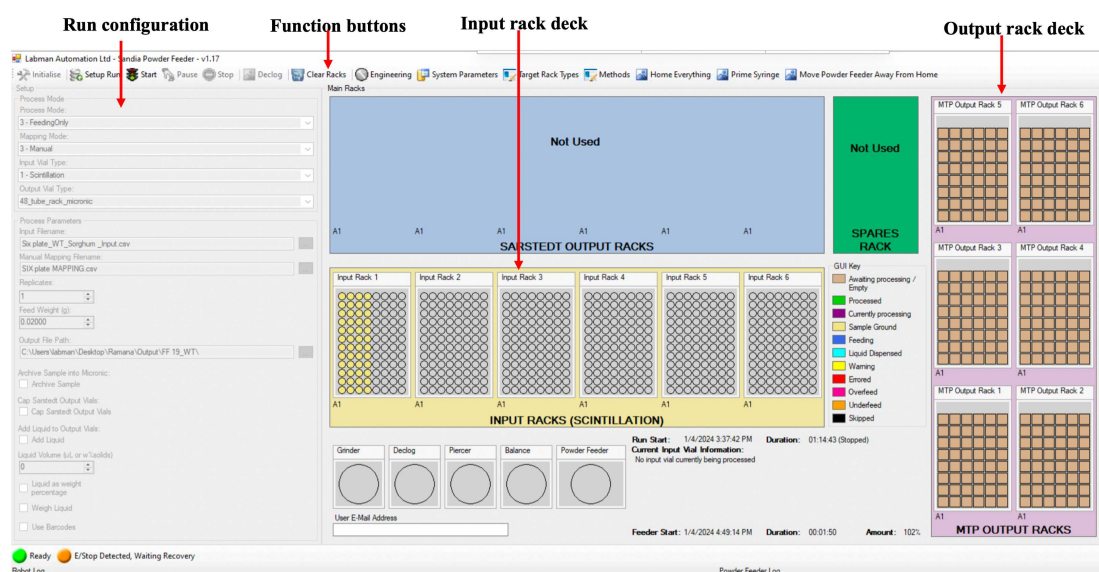
Run Configuration - Process Mode (top left corner)

`1m`

2 **Process Mode:** - Select "FeedingOnly"

`1m`

Mapping Mode - Select "Manual"

Input Vial Type - Select "Scintillation tube" or "Sarstedt tube" depending on the format of your samples

Output Vial Type - Select "Micronic 48"

Run Configuration - Process Parameters (middle left side)

`5m`

3

Process Parameters:

Input Filename - Input csv file
Manual Mapping Filename - manual mapping csv file
Output File Path - Output file path
Input filename - The input file ('Input Filename' option) is a csv file that tells the robot where the source vials will be located. It has four columns.

A	B	C	D
Sample Description	Source Rack Number	Source Vial position	Method Name
1	1	1	200mg_sorghum_method
1	1	2	200mg_sorghum_method
1	1	3	200mg_sorghum_method
1	1	4	200mg_sorghum_method
1	1	5	200mg_sorghum_method
1	1	6	200mg_sorghum_method
1	1	7	200mg_sorghum_method
1	1	8	200mg_sorghum_method
1	1	9	200mg_sorghum_method
1	1	10	200mg_sorghum_method
1	1	11	200mg_sorghum_method
1	1	12	200mg_sorghum_method
1	1	13	200mg_sorghum_method
1	1	14	200mg_sorghum_method

Column A - Required, do not modify.
Column B - This column indicates the rack position for the source vials. These numbers are indicated on the deck.
Column C - This is the source well of the tube rack indicating where the source well is placed. 1 = A1 (bottom left corner), and 96 = H12 (top right corner).
Column D - This is the dispensing method that will be used by the Labman for distributing biomass.

4   **Manual Mapping filename** - The "Manual Mapping" file is a csv file that assigns a source (or input) vial to

a destination (or output) vial. Note that these columns have labels, while the input file does not.

A	B	C	D	E	F
OutputRack	OutputRackPosition	InputRack	InputRackPosition	DispenseWeight (g)	LiquidVolume (uL)
1	1	1	1	0.2	0
1	2	1	1	0.2	0
1	3	1	1	0.2	0
1	4	1	1	0.2	0
1	5	1	1	0.2	0
1	6	1	1	0.2	0
1	7	1	1	0.2	0
1	8	1	2	0.2	0
1	9	1	2	0.2	0
1	10	1	2	0.2	0
1	11	1	2	0.2	0
1	12	1	2	0.2	0
1	13	1	2	0.2	0
1	14	1	2	0.2	0
1	15	1	3	0.2	0

Column A - OutputRack → This is the rack position where the destination plates will sit. For micronic48 tube racks, these will sit in the "MTP Output Rack" position on the deck (see Software Layout figure for details).

Column B - OutputRackPosition → This is the well in the tube rack that contains the destination tube used for this step. Numbering and position of wells is the same as for the input csv file (A1 = 1, F8 = 48, A1 is at bottom left hand corner if facing the robot).

Column C - InputRack → This is the deck position where the source tubes are located. This should be the same as the rack specified in "Column B" of the Input CSV file.

Column D - InputRackPosition → This is the well location of the rack where the source tube will be located. Note that numbering and layout convention is the same as used for the Input file CSV.

Column E - DispenseWeight → The weight (in g) of biomass to be transferred from the source tube to the destination tube.

Column F - LiquidVolume → Although we NEVER use liquid dispensing on the Labman, this column

must be specified on the csv file, and should always be set to 0.

5   **Output file path** - Create a new file prior to each run with the date in the name.

> **Note**
>
> The output files specify information such as intended vs actual amount of biomass distributed, and if any error occurs. For every well completed, a new csv file is specified containing all completed wells. The address located here is where output files are specified.

Starting a Run   5m

6   Setting up the deck:

1. Place all source and destination tubes on the deck in the designated positions.
2. Once the deck is set, close the doors completely. They are all magnetic, and should click into place.
3. Then click the big green button - located next to the FRONT emergency stop button.
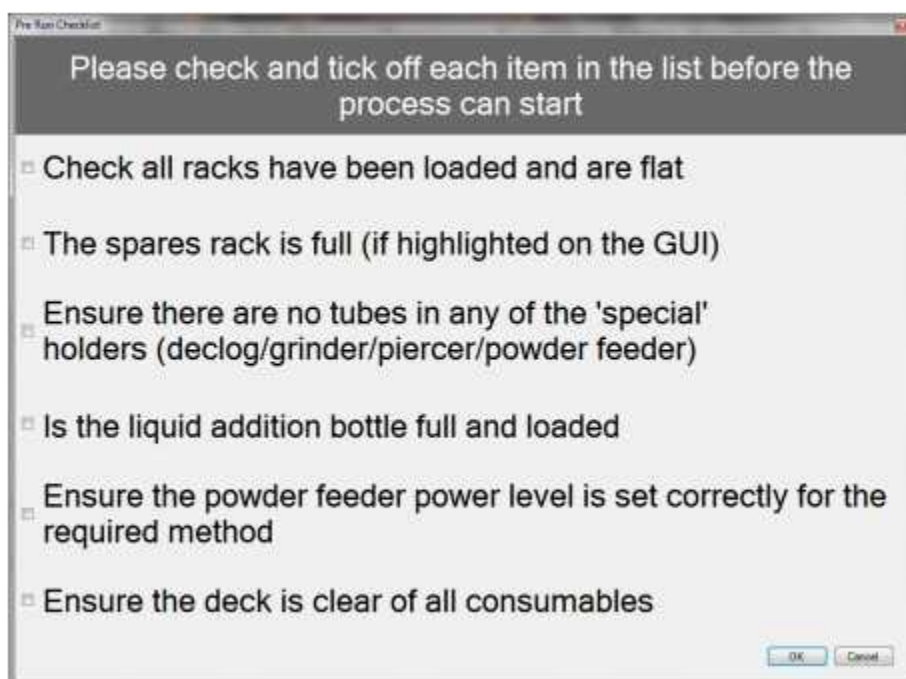
> **Note**
>
> - The doors have a pneumatic trigger that freezes all activity when opening. Thus, you might hear a "hiss" when you open the doors.
>
> - If it is closed well, there should be another audible "hiss", indicating that the robot is ready to run.

7   Click the "**Setup Run**" button in the "Function Buttons" portion of the screen.

> **Note**
>
> If the specified csv files are correct, then the well positions containing source and destination tubes should be highlighted in a beige color.

8   Once this is complete, click the "**Start**" button of the "Function Buttons". You should see the following checkbox:

Note

This box simply exists as a reminder of safe operation to the user. All boxes must be checked prior to hitting OK even if that feature is not being used.

Clicking on 'OK' will initiate the run.

Note

It takes ~ 1 min to distribute ~200 mg of biomass into each micronic48 tube. This means that each full micronic plate will take ~ 1hr to complete a full plate.