



HYDRA USAGE

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SUMMARY

This protocol outlines the procedure of maintaining and using the Robbins Scientific Hydra.

MATERIALS & REAGENTS

<u>Materials/Reagents/Equipment</u>	<u>Vendor</u>	<u>Catalog Number</u>
REAGENTS		
2% Bleach	Homemade	
Coulter Clenz	Fisher Scientific	23-257520

EQUIPMENT

Hydra	Robbins Scientific	
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PROCEDURE

Preparation for Usage

1. First thing in the morning wash the Hydra. Press red set/reset button until the LED display reads D...x (This step is important or else it will not wash properly). Then set it to FILE 1 and use the reservoir filled with Coulter Clenz solution. Note: Change the Coulter Clenz solution weekly. Place the reservoir filled with Coulter Clenz on the dispensing platform. Push the WASH button and come back later. It will take about 10-15 minutes to finish the wash.
2. Remove the Coulter Clenz reservoir and place a reservoir filled with water on the dispensing platform. Push the WASH button and again come back later. Repeat this step one more time.
3. Now the hydra is ready for use.

Usage

1. Be careful when using the Hydra, it is easy to crash the needles with the press of a wrong button or on the wrong file. Always pay attention as a distracted mind can get you in trouble.
2. When the LED display on the front top of the unit reads D...x, the Hydra is ready for usage and you can now access the first file of your procedure. D refers to the current file's mode (Dispense) and x refers to the current volume of solution in the syringes.
Note: If the display reads EMPTY, put a reservoir on the dispensing platform and press the EMPTY button. This will empty out the solution in the syringes into the reservoir.
3. Push the set/reset button to move through all the parameter settings in the file.
4. Verify all the file-parameter settings of files that you will use in the protocol.
5. Follow the protocols instructions.

6. When you have finished all the procedural steps, including the Wash operation, empty the syringes by placing a reservoir on the dispensing platform and pressing the EMPTY button.
Note: Only use 2% bleach when the protocol calls for it, never use it to wash the syringes as salt build up can form and may inhibit some of our reactions.
7. When you are through using the Hydra for the day, replace the wash solution with fresh water for one final additional wash cycle on FILE 1. Before you press the WASH button, position yourself so you can easily reach the power switch on the back of the Hydra. Press WASH, and when the syringes are full and the dispensing platform is up so that the needle tips are submerged by at least 2 cm, press the power switch to OFF. Now both the needle tips and the interior of the syringe barrels will be bathed in the water during the storage interval.

Overview of Hydra Files

FILE 1: This file is used for daily washing.

FILE 2: This file is used for 96 well *E-coli* plates. This file is used for **RCA setup** using the Hydra. It can fill from a 96 well U bottom plate and dispenses into a full skirted 96 well plate.

FILE 3: This file is used to fill from a 384 well GENETIX plate and dispenses into an U-bottom 96 well plate.

FILE 4: This file is used to fill from a half skirted 96 well plate if needed and dispenses into a half skirted 96 well plate.

FILE 5: This file is used to fill full skirted 96 well plate and dispense into full skirted 96 well plate. This file is usually used when aliquoting solutions into 96 well plates.

FILE 6: This file is used to fill from regular 384 well plates and dispense into half skirted 96 well plate. This file is usually used when transferring post-sequencing reaction from 384 well plate into half skirted 96 well plate for Ethanol Precipitation.

FILE 7: This file is used to fill full skirted 96 well plate and dispense into half skirted 96 well plate. This file is usually used when transferring pre-sequencing reaction mixture into half skirted 96 well plate to run it on the thermal cyclers.

FILE 8: This file is used to fill full skirted 96 well plate and dispense into regular 384 well plate. This file is usually used when transferring pre-sequencing reaction mixture into regular 384 well plates to run it on the thermal cyclers.

Wash Procedures

For *E-coli* Samples: Fill a reservoir with 2% bleach. After going through the RCA setup steps empty the rest of the *E-coli* into the reservoir filled with 2% bleach. Fill another reservoir with fresh water and press WASH in FILE 4. This should go through 3 wash cycles at a wash height of 20. Then replace the wash solution with fresh water and press WASH again. Finally, grab another reservoir and fill it with fresh water for one more final wash, press the WASH button.

For Amplified DNA Samples: Fill a reservoir with fresh water and press WASH in which ever file is being used, this should go through 3 wash cycles at a wash height of 20. Then replace the wash reservoir with a new one filled with fresh water, press WASH. After the wash set aside the reservoir filled with the dirty water so that you can empty the excess DNA sample into this reservoir and then toss out the water. Repeat wash steps.