

# EVUSEP

**Sample loading  
protocol for Evotips**



# Evotips

Evotips are disposable, purification and loading trap columns.

Evotips fit most standard pipettes and commercial robotic systems.

Using Evotips results in simplified workflows that removes several sample handling steps and reduces injection cycle overheads.

Carry-over is further minimized by eluting with a partial gradient and in a very precise and reproducible volume that leaves impurities and high-molecular-weight contaminants behind.

## Technical details

| Specification         | Value   |
|-----------------------|---|
| Volume                | 0.5-200 $\mu$ l   |
| Pore size             | 100 Å   |
| Max. loading capacity | 1 $\mu$ g of complex peptide mixture or 1 pmol of simple protein digest |



## Ordering information

| Part Number | Description                       |
|-------------|-----------------------------------|
| EV2001      | Evotip C18, 1 box (96 tips)       |
| EV2003      | Evotip C18, 10 boxes (960 tips)   |
| EV2005      | Evotip C18, 50 boxes (4800 tips)  |
| EV2008      | Evotip C18, 100 boxes (9600 tips) |

## Required Material

1. Box of Evotips
2. Mass spec grade 1-propanol, alternatively 2-propanol
3. Solvent A: Mass Spec Grade Water with 0.1% Formic Acid, JT Baker P/N: JT-9834-2 or equivalent
4. Solvent B: Mass Spec Grade Acetonitrile with 0.1% Formic Acid, JT Baker P/N: JT-9832-2 or equivalent
5. Multichannel pipette
6. Solvent containers for multichannel pipette, e.g. Dual Solution Reservoir HEA20281A from Heathrow Scientific [www.heathrowscientific.com/dual-solution-reservoir-i-hea20821a](http://www.heathrowscientific.com/dual-solution-reservoir-i-hea20821a)
7. 96 well Microtiter plate (MTP), e.g. 249946, 96 well Conical Btm PP Plt Natural from Thermo Scientific, [www.thermofisher.com/order/catalog/product/249946](http://www.thermofisher.com/order/catalog/product/249946)
8. Lids for 96 well MTP, e.g. 263339 Nunc™ Microplate Lids, [www.thermofisher.com/order/catalog/product/263339](http://www.thermofisher.com/order/catalog/product/263339)
9. Evotip adapter rack (EV1068)
10. Gloves
11. Table centrifuge
12. Fume hood

## Important info

- **At all centrifugation steps use max acceleration and deceleration**
- **After all centrifugation steps, empty the box for solvent**
- **Please note that following settings are guidelines, adjustment might be needed depending on centrifuge.**
- **Centrifugation should be room temperature**

**Wear safety glasses, coat and gloves and work in fume hood when following this procedure.**

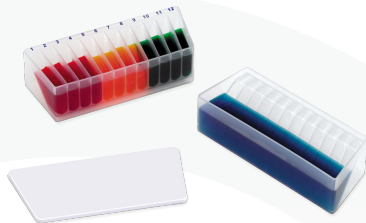
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## Prepare solvents and containers

1. **Pour Solvent A, Solvent B, and 1-propanol into their respective solvent containers.**

(The recommended dual purpose containers can either be used as a single reservoir or as 12 individual ones).

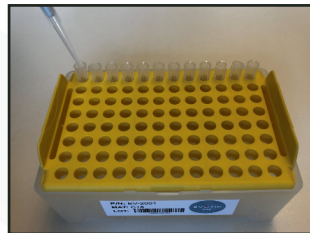
2. **Pipette 100  $\mu$ l of 1-propanol into each well of a 96 well plate MTP using the multichannel pipette.**



## Wash

1. Transfer 20  $\mu$ l Solvent B to all Evotips
2. Place the Evotips in the centrifuge with appropriate counter balance and centrifuge 700 g for 60 seconds

**REMEMBER TO EMPTY BOX  
FOR SOLVENT B AFTER  
CENTRIFUGATION STEP**

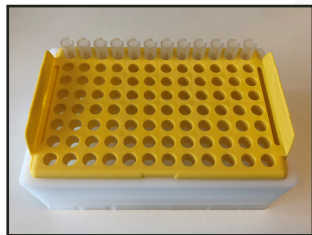


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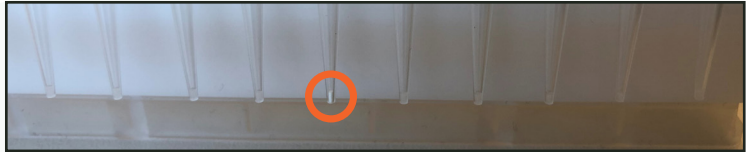
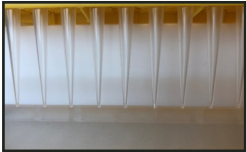
## Condition


1. Place the Evotip adaptor rack on top of the MTP with 1-propanol
2. Soak for a minimum of 10 seconds and then visually inspect that tips are pale white

**VERY IMPORTANT THAT ALL TIPS  
ARE PALE WHITE AFTER SOAKING**



# Let the tips soak until all are pale white



 *Not pale white*

*Pale white vs white*

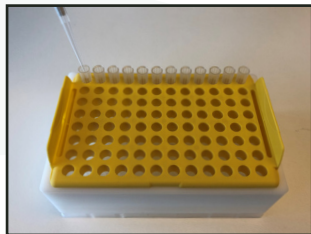
**This step might take  
up 30-60 seconds**



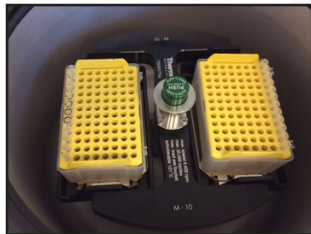
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## Equilibrate

With the Evtips still in the adapter rack, transfer 20  $\mu$ l Solvent A to each Evtip



Place the Evtips in the centrifuge with appropriate counter balance and centrifuge 700 g for 60 seconds





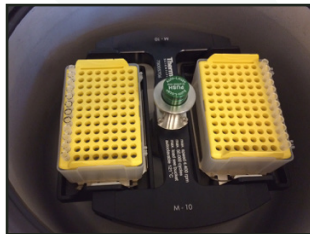
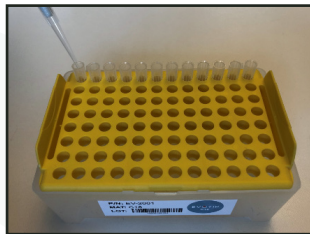
## Sample load

Place the tips in the original tray and transfer the sample (20  $\mu$ l) to each Evtip

**DO NOT PLACE THE TIPS IN THE  
1-PROPANOL ADAPTOR AT THIS STAGE!**

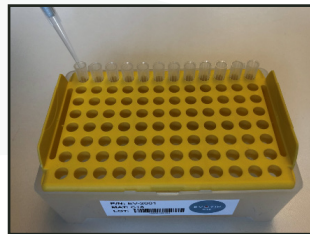
Transfer Evtips to Solvent A tray, if loading time exceeds a few minutes to keep Evtips wet

Place the Evtips in the centrifuge with appropriate counter balance and centrifuge 700 g for 60 seconds



# Wash

Transfer 20  $\mu$ l solvent A to each EvoTip



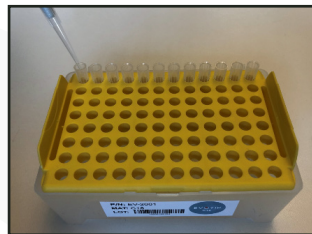
Place the EvoTips in the centrifuge with appropriate counter balance and centrifuge 700 g for 60 seconds



## Preservation

Transfer 100  $\mu$ l Solvent A to each Evotip

Place the Evotips in the centrifuge with appropriate counter balance and centrifuge 700 g for 10 seconds to keep tips wet



## Storage recommendation

Add enough solvent A to the bottom of the tray so the bottom of the Evotips are submerged and store cold with lid on until analysis



# Quick guide



## WASH

Wash dry tips with 20  $\mu$ l solvent B and centrifuge for 60 s at 700 g



## CONDITION

Soak in 100  $\mu$ l 1-propanol until all the tips are pale white



## EQUILIBRATE

Equilibrate soaked tips with 20  $\mu$ l solvent A and centrifuge for 60 s at 700 g



## LOAD

Load samples on wet tips (20  $\mu$ l in solvent A) and centrifuge for 60 s at 700 g



## WASH

Wash tips with 20  $\mu$ l solvent A and centrifuge for 60 s at 700 g



## WET

Transfer 100  $\mu$ l solvent A and centrifuge tips for 10 s at 700 g to keep tips **wet**



# EVOSEP

## NOTES

1. Ensure Evtips are continuously wet after soaking in 1-propanol
2. Transfer Evtips to solvent A tray in loading step to avoid them drying out
3. Prepare Evtips just prior to MS analysis, but for longer storage add solvent A in the bottom of the box and store cold with lid on

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