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We use this protocol and it's working

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# Extraction of 1-HP derivatives and HPLC preparation

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ABSTRACT

This protocol describes day 6 of our workflow to grow worms in liquid culture to induce the production of natural products (in this case, 1-HP derivatives). In this protocol, we extract 1-HP and its derivatives and then prepare the sample for HPLC injection.

1 Remove the sample from the lyophilizer. It should appear to be a powder.

2 Resuspend in minimal methanol (enough to submerge the pellet)

3 Vortex the tube with methanol to aid in dissolution.

4 The HPLC injection sample caps out at 90 uL, so minimizing the solution volume here will minimize the total number of injections needed.

5 Transfer the solution to a 2 mL microcentrifuge tube. Centrifuge at 20,800 RCF for 30 minutes.

6 Transfer only the supernatant into HPLC tubes (leave out the solids if any at the bottom of the microcentrifuge tubes). Keep track of the volume so that the injection volume for HPLC will be accurate.

7 Place HPLC tubes in the correct loading sites of the machine and run the protocol.

8 While the HPLC is ongoing, collect fractions if desired (either manually or automatically). Once collected, fractions can be stored at -80˚C for NMR prep later.