LSOP Title	Arabidopsis growth on agar plates
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Version	1.1
Location	UQ Node/Centre-wide
Policy/Procedure Link	*UQ- Equipment* *OGTR*
Risk Assessments	
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1. Scope

To outline the procedures for growing Arabidopsis on agar for CK extraction

This LSOP does not cover the growth of Arabidopsis in soil or any other medium. It also does not include how to create agar or agar plates.

2. Definitions

EtOH – ethanol

Murashige and Skoog (MS) – a type of selective medium

3. Materials and Equipment

1. Square petri dishes
2. MS agar
3. Sucrose
4. Ethanol
5. Pipette
6. Microcentrifuge tube
7. Distilled water
8. Alfoil

9. Laminar Flow

4. Prescribed Actions

1. Prepare square petri dishes with 50 mL half-strength Murashige and Skoog (MS) medium containing 1% sucrose and 1% agar (pH 5.7).

   NB: autoclave 20% sucrose solution and MS agar separately, then add sucrose to the MS medium prior to pouring the plates.

   NB: you can create any media you require for this step

2. Sterilise Arabidopsis seeds (LSOP05)
3. Dispense seeds to be used into a new sterile microcentrifuge tube and add ~200 uL of sterile distilled water or 2% agar (see appendix or LSOP05 appendix)
4. Using a pipette, suck up water/agar until you get a single seed in the pipette tip, then dispense the liquid + seed onto the plate. Place seeds in a line across the center of the plate. Repeat until you have dispensed ~15-20 seeds per plate across all plates.
5. Wrap plates in alfoil and stratify by keeping at 4°C for two days.

   N.B. This should be done with plates flat.

6. Places plates upright (with the line of seeds horizontal) in a growth cabinet and grow for 4 weeks at desired growth conditions (i.e. 150, 25^{o}C, 12h days).

5. Appendix

0.2% Agar

1. In a small Schott bottle, add 0.2 g agar to 100 mL milliQ water.
2. Autoclave (LSOP83).
3. Add ~ 1.5 mL (depending on number of seeds) sterile 0.2% agar solution to Arabidopsis seeds in Eppendorf to vernalise / plate