

Temperature Gradient for WPS2 Plates

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Since previous experiment (80 C, 5') showed no fungal grows on the SA/SAL plates

Proposed the matrix of conditions for WPS2 enrichment - tentative spore former:

Dilutions 1:1000, 1:10000 should be considered - since they showed optimal in the previous run

| | 1' | 2' | 3' | 4' | 5' |
|--------|----|----|----|----|----|
| A:55 C | A1 | A2 | A3 | A4 | A5 |
| B:65 C | B1 | B2 | B3 | B4 | B5 |
| C:75 C | C1 | C2 | C3 | C4 | C5 |
| D:85 C | D1 | D2 | D3 | D4 | D5 |

! (Do plate pipetting from back to front: assuming that 1' is the least important)

! Later maybe do the same with liquid enrichments

Number of samples, media requirements:

| | No | ml |
|-----------|----|-----------|
| SA | 20 | 20x25=500 |
| SAL | 20 | 20x25=500 |
| 1:1000:A | 5 | 5*200u=1 |
| 1:10000:A | 5 | 5*200u=1 |
| 1:1000:B | 5 | 5*200u=1 |
| 1:10000:B | 5 | 5*200u=1 |
| 1:1000:C | 5 | 5*200u=1 |
| 1:10000:C | 5 | 5*200u=1 |
| 1:1000:D | 5 | 5*200u=1 |
| 1:10000:D | 5 | 5*200u=1 |

Soil Agar (SA)/ Soil Agar L. (SAL)

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Soil Agar SA 500 ml

Tap water

(Medium pH: ~4.7 <before autoclaving>)

| | | |
|---------|-----|----|
| H2O | 500 | ml |
| Soil | 50 | g |
| Agarose | 7.5 | g |

Soil Agar L. SAL 500 ml

- Original protocol uses sieved soil: Use just ground in the mothur
- Distilled water

(Medium pH: ~4.7 <before autoclaving>)

| | | |
|-------------------|-------------------------------|--------------|
| dH2O | 400 | ml |
| K2HPO4 | 0.1 (Not doing it) | g |
| Soil | 350 | g |
| Agarose | 7.5 | g |