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Complete Medium or Complete Medium Xylose (from Leach, Lang and Yoder 1982) V.2

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Protocol status: In development

We are still developing and optimizing this protocol

Created: May 16, 2023

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Abstract

For the growth and maintenance of *Cochliobolus carbonum* and *Cochliobolus victoriae*

Materials

Trace Minerals

Boric Acid (H₃BO₃) CAS: [10043-35-3](#)

Cupric Sulfate (CuSO₄) CAS: [7758-98-7](#)

Potassium Iodide (KI) CAS: [7681-11-0](#)

Manganese (II) sulfate monohydrate MnSO₄ * H₂O CAS: [10034-96-5](#)

Sodium permanganate monohydrate NaMoO₄ *H₂O CAS: [79048-36-5](#)

Zinc sulfate heptahydrate ZnSO₄ * 7 H₂O CAS: [7446-20-0](#)

Iron (III) chloride hexahydrate FeCl₃ * 6H₂O CAS: [10025-77-1](#)

Salts

Calcium nitrate tetrahydrate Ca(NO₃)₂ * 4 H₂O CAS: [13477-34-4](#)

Potassium Phosphate monobasic KH₂PO₄ CAS: [7778-77-0](#)

Magnesium Sulfate heptahydrate MgSO₄ * 7H₂O CAS: [10034-99-8](#)

Sodium Chloride NaCl CAS: [7647-14-5](#)

Media

Yeast Extract CAS: 8013-01-2 (some people report differences between different sources)

Casein Digests: Still a bit of work to do here to determine, which of these is best (including new plant-based sources).

Big differences in prices between different digest types.

Acid hydrolysed Casein CAS: [65072-00-6](#)

Peptone from casein, tryptic digest OR pancreatic digest (tryptone) CAS: [91079-40-2](#)



Make Micronutrients Solution

- 1 9 mg H_3BO_3
58.5 mg $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$
1.95 mg KI (Potassium Iodine)
9 mg MnSO_4
7.6 mg NaMoO_4
822 mg $\text{ZnSO}_4 \cdot 6\text{H}_2\text{O}$
139.8 mg $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$

in 300 mL ddH₂O and filter sterilise

Citation:

Heterokaryosis and Parasexuality in the Fungus *Ascochyta Imperfecta* Author(s): K. E. Sanderson and A. M. Srb Source: *American Journal of Botany*, Jan., 1965, Vol. 52, No. 1 (Jan., 1965), pp. 72-81 Published by: Wiley Stable URL: <https://www.jstor.org/stable/2439977>

Make 100x Salt Solutions A and B

- 2 **100X Salt Solution A**

10g $\text{Ca}(\text{NaO}_3)_2 \cdot 4\text{H}_2\text{O}$
100 mL ddH₂O

Autoclave

Citation:

Leach, J., Lang, B. R. & Yoder, O. C. *Microbiology***128**, 1719-1729, [doi:https://doi.org/10.1099/00221287-128-8-1719](https://doi.org/10.1099/00221287-128-8-1719) (1982).

- 3 **100X Salt Solution B**

2 g $\text{KH}_2\text{PO}_4 \cdot 7\text{H}_2\text{O}$
1.5g NaCl

100 mL H₂O
pH 5.3

Autoclave



Citation:

Leach, J., Lang, B. R. & Yoder, O. C. *Microbiology***128**, 1719-1729,
[doi:https://doi.org/10.1099/00221287-128-8-1719](https://doi.org/10.1099/00221287-128-8-1719) (1982).

Make Complete Medium (CM) or Complete Medium Xylose (CMX)

4 Complete Medium Base

10 g glucose OR xylose (substitute glucose for xylose for CMX medium)
1 g Yeast Extract
0.5 g Acid-hydrolysed Casein
0.5 g Enzyme-hydrolysed casein
20g Agar
10 mL Salt A
10 mL Salt B

Make up to 1000 mL with ddH₂O and Autoclave

After autoclaving add:

1 mL sterilised micronutrient solution

Citation:

Leach, J., Lang, B. R. & Yoder, O. C. *Microbiology***128**, 1719-1729,
[doi:https://doi.org/10.1099/00221287-128-8-1719](https://doi.org/10.1099/00221287-128-8-1719) (1982).

Complete Medium for Sporulation

5 Complete Medium Base

0.5 g glucose OR xylose (substitute glucose for xylose for CMX medium)
20 g Sorbose
1 g Yeast Extract
0.5 g acid hydrolysed casein
0.5 g enzyme hydrolysed casein
20g Agar

10 mL Salt A
10 mL Salt B

Make up to 1000 mL with ddH₂O and Autoclave

After autoclaving add:

1 mL sterilised micronutrient solution

Protocol references

Citation:

Heterokaryosis and Parasexuality in the Fungus *Ascochyta Imperfecta* Author(s): K. E. Sanderson and A. M. Srb Source: *American Journal of Botany* , Jan., 1965, Vol. 52, No. 1 (Jan., 1965), pp. 72-81 Published by: Wiley Stable URL: <https://www.jstor.org/stable/2439977>

Leach, J., Lang, B. R. & Yoder, O. C. *Microbiology***128**, 1719-1729, [doi:https://doi.org/10.1099/00221287-128-8-1719](https://doi.org/10.1099/00221287-128-8-1719) (1982).