Chaima Denhadji
Fungal Biodiversity and Biotechnology Lab-
UMR 1163 INRAE - AMU
France, Marseille

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Intership finance by AGRIMA Erasmus+

Impact of the Endogenus Flora on the Development of *Polyporus brumalis* during SSF on Miscanthus

Summer Camp | July 7 – 11, 2025 | Marseille, France

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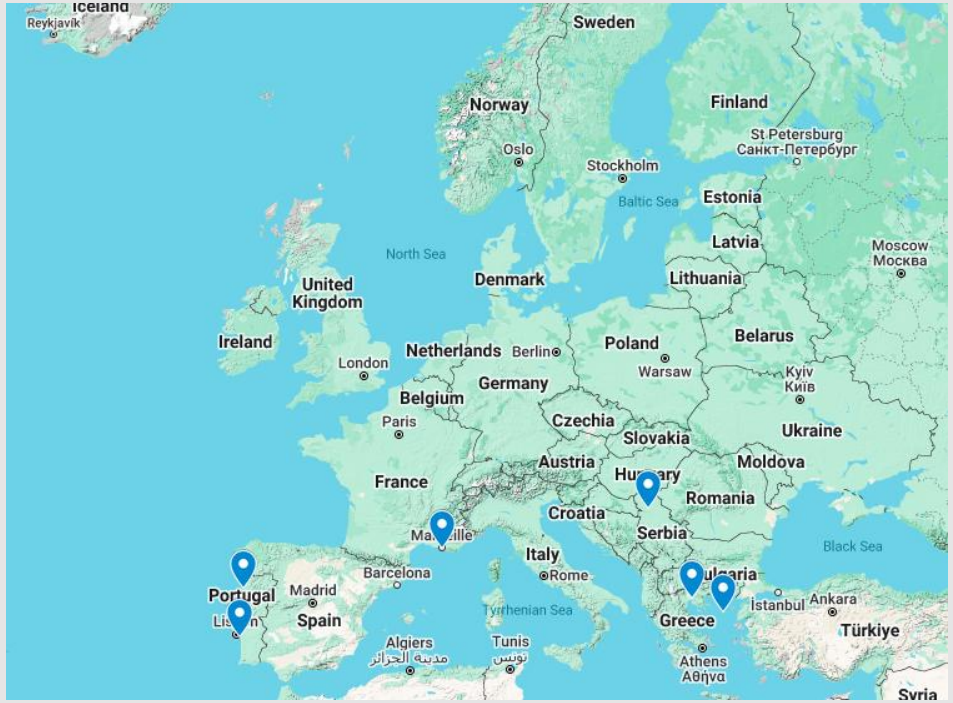

amU Aix Marseille Université

AVIPE Associação de Viticultores do Concelho de Palmela

P.PORTO

UNIVERSITAS STUDIORUM NEOPLANTENSIS UNIVERSITY OF NOVI SAD

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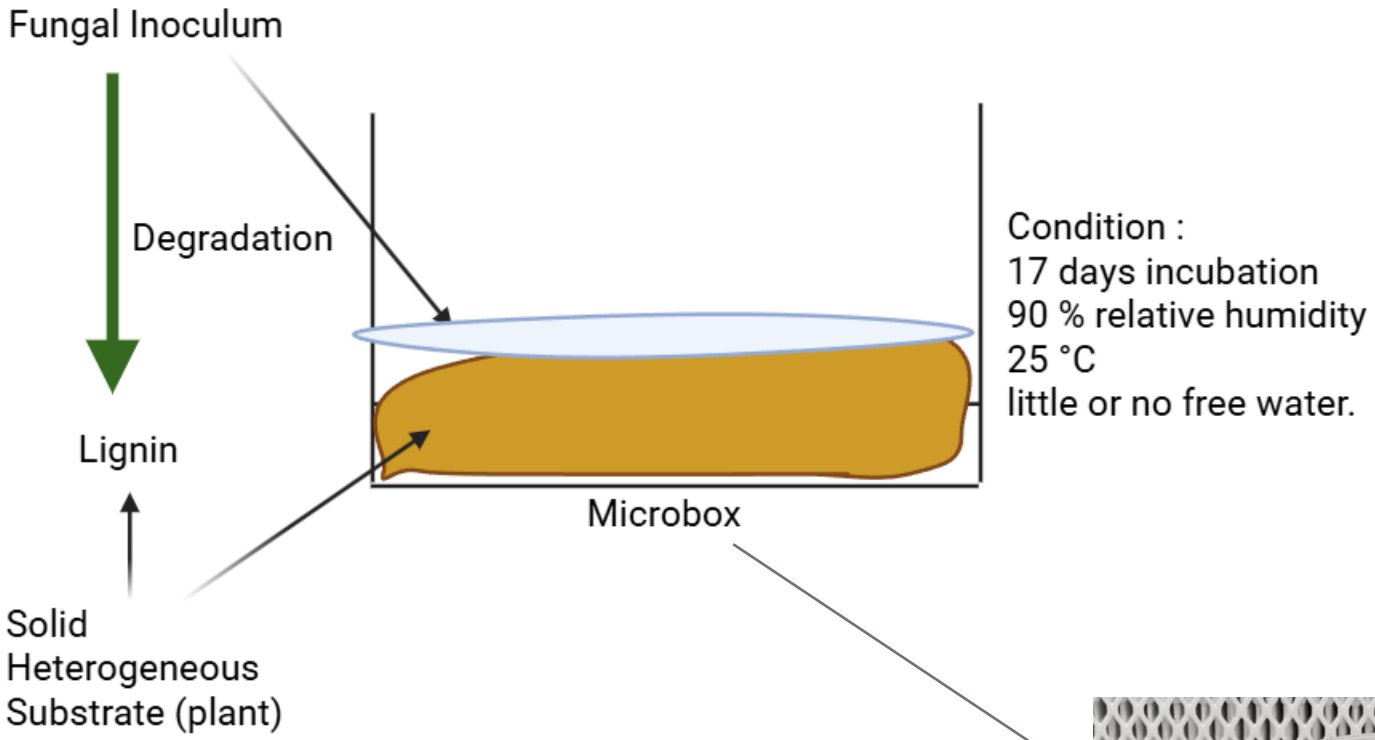
Solid-State Fermentation (SSF)



BENEFITS:

- Adapted to filamentous fungi on plant biomass
- Low volumes of water
- Agricultural waste : low-cost nutrients for microorganisms

https://www.lesaffre.com/fr/trends-mag/fermentation-solide-des-applications-porteuses-davenir/#:~:text=La%20fermentation%20sur%20substrat%20solide%20est%20un%20proc%C3%A9d%C3%A9%20durant%20lequel,est%20pas%20soluble%2C%20mais%20solide.

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A limitation for SSF at industrial scale is the cost of decontamination

Test if the presence of endogenous flora is compatible with the establishment of the fungus on plant biomass in an SSF process

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Miscanthus

https://www.francebleu.fr/emissions/terre-de-champagne-ardenne/qu-est-ce-que-le-miscanthus-comment-est-il-cultive-et-pour-quels-usages-7540363

The stem ~ 75-80% holocellulose + 25-30% lignin

- Grown in France
- Renewable resource
- NO Food use
- NO Much water
- NO Pesticides or fertilizers

https://france-miscanthus.org/wp-content/uploads/2017/12/Fiche_culture_RMT_Biomasse_2009.pdf

https://docnum.univ-lorraine.fr/public/SCD_T_2010_0063_EL_HAGE.pdf

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Polyporus brumalis (White-rot fungus)

Xylanases

Hemicellulose Degradation

Mannanases

Enzymes → Cellulases → Cellulose Degradation

Class II Peroxydases

Lignin Degradation

CIRM,BBF

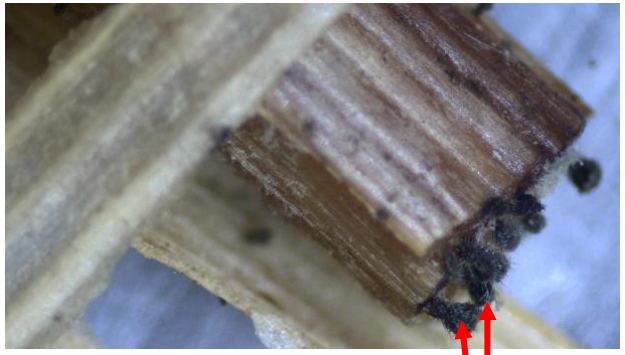
https://www.researchgate.net/figure/Cellulose-hydrolysis-by-the-three-types-of-cellulases-enduglucanase-breaks-the_fig1_236088767

1/ Is *P. brumalis* able to develop on non-sterilized biomass?

Microscopic observation of sterilised and non-sterilised miscanthus

Non-Sterilized Plant Substrate

Microscopic Observation x51



Endogenous Microorganisms

Sterilized Plant Substrate (110°C 30 min)

Microscopic Observation x51

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Non-Sterilized and Sterilized Miscanthus with Fungal Inoculum (30 mg dry mass of mycelium / g of dry mass miscanthus)

	Sterilized miscanthus + fungal inoculum	Non-sterilized miscanthus + fungal inoculum
Day 15		

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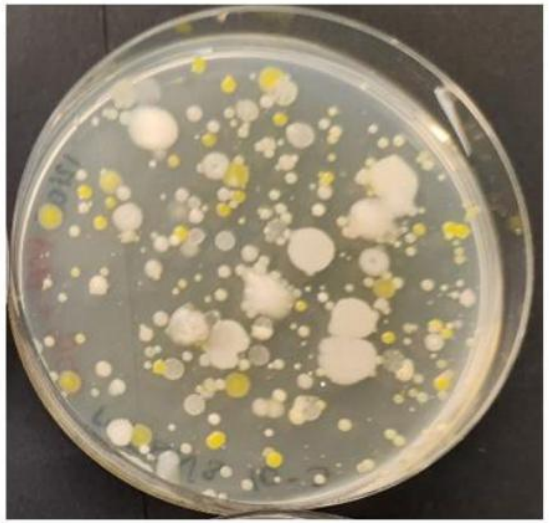
2/ Does *P. brumalis* impact the endogenous bacteria and fungal flora ?

Analysis of the Bacterial Flora of Miscanthus on PCA (Plate Count Agar)

A Non-sterilized miscanthus + fungal inoculum (PCA)

B Non-sterilized miscanthus + fungal inoculum (PCA + Cycloheximide = inhibits fungal growth)

Days 3



The bacterial population is observable

Macroscopic observation,3 days incubation at 25°C, on PCA (A) and PCA + Cycloheximide (B)

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Analysis of the Fungal Flora of Miscanthus in Sabouraud + chloramphenicol medium

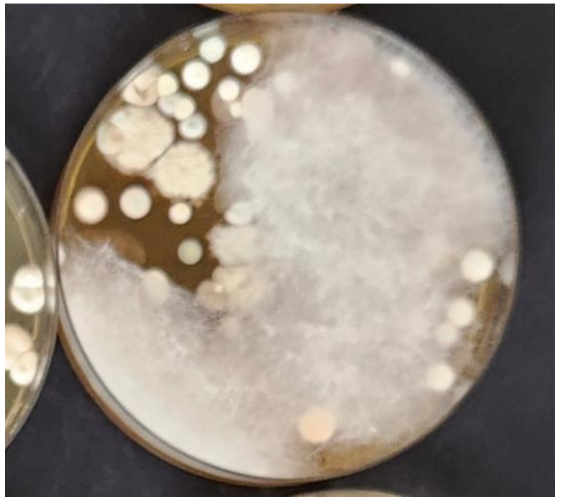
A

Non-sterilized miscanthus

B

Non-sterilized miscanthus + fungal inoculum

Days 5



Macroscopic Observation, Incubation at 25°C

In conclusion, whether inoculated or not = presence of fungal diversity

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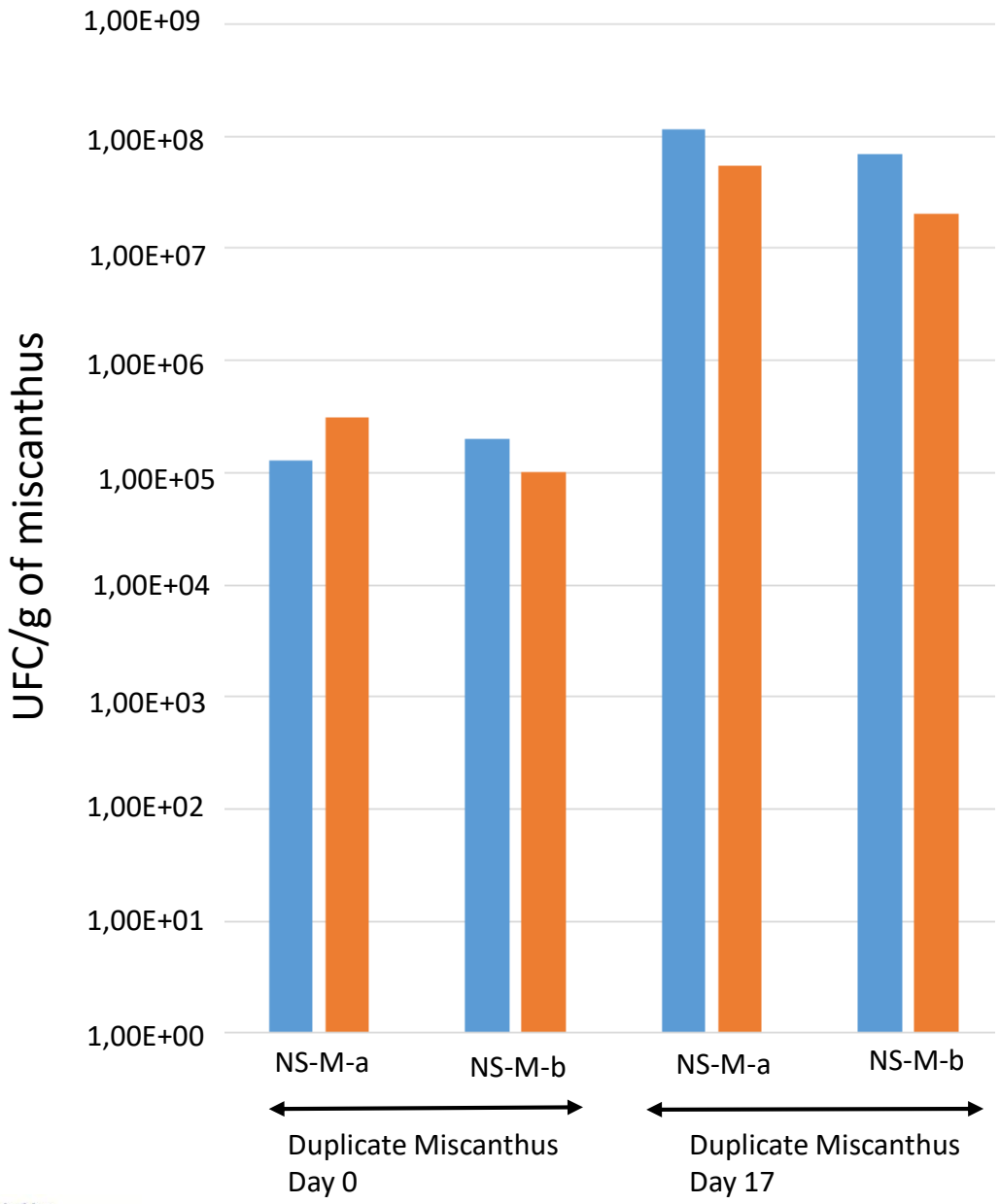
Evolution of the Fungal and Bacterial Population over Time



Endogenous Bacterial Flora

Endogenous Fungal Flora

Non-sterilized Miscanthus : NS-M

25°C
90 % Relative humidity
17 day incubation

After 17 days of incubation

↓

↑ **endogenous flora.**

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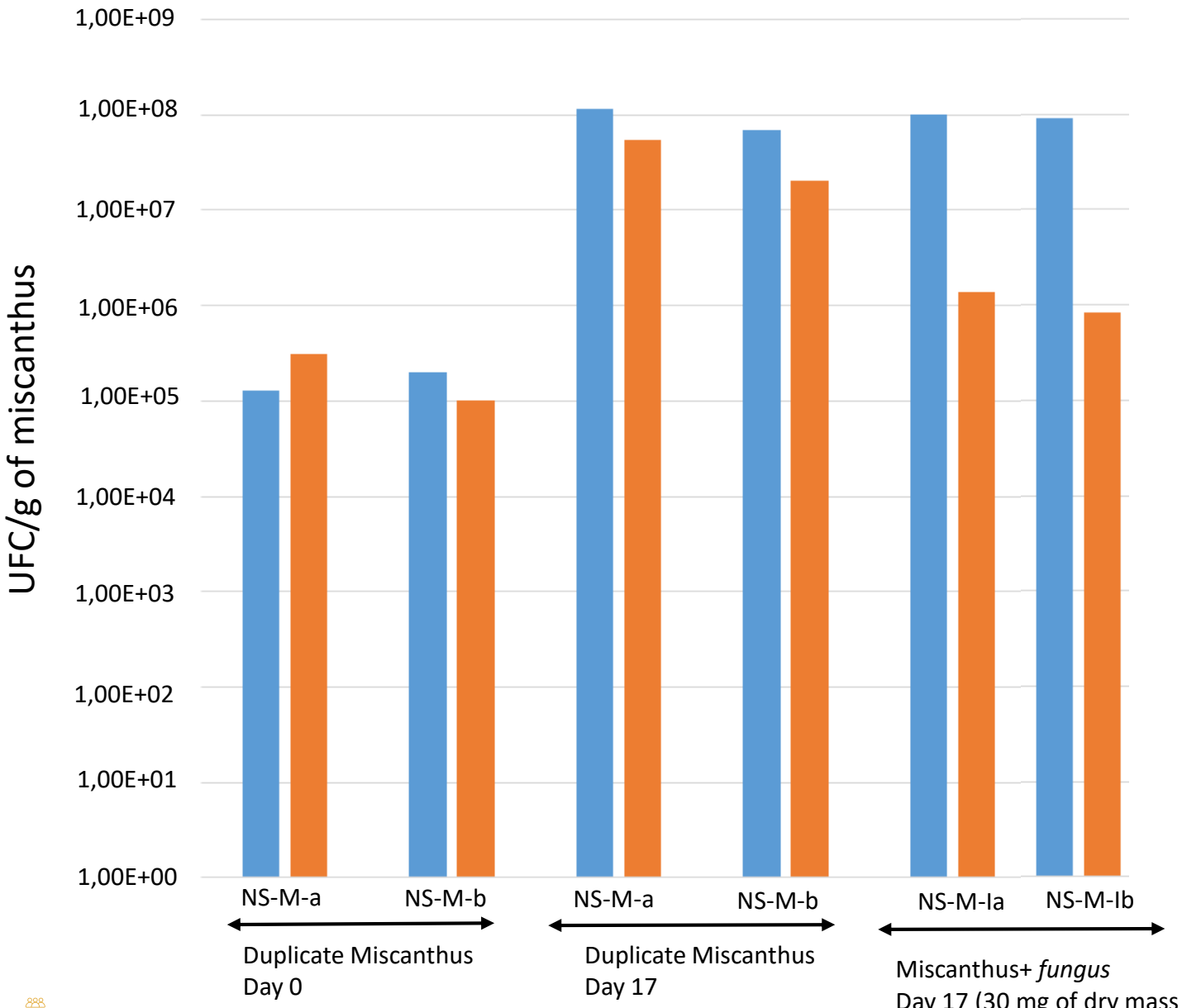
Evolution of the Fungal and Bacterial Population over Time in the Presence of the Fungus



Endogenous Bacterial Flora

Endogenous Fungal Flora

Non-sterilized Miscanthus : NS-M
Non-sterilized Miscanthus Inoculated with the fugus: NS-M-I

25°C
90 % Relative humidity
17 days incubation

In the presence of the fungus: no effect on the bacterial population but negative effect on the fungal population

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Macroscopic observation with different inoculation rates of *P. brumalis* (after 15 days)

	Non-sterilized Inoculated miscanthus	Sterilized Inoculated miscanthus
30 mg of dry mass mycelium / g of dry mass Miscanthus		
10 mg of dry mass mycelium / g of dry mass miscanthus		

Even with a low level of inoculum, the fungus grows

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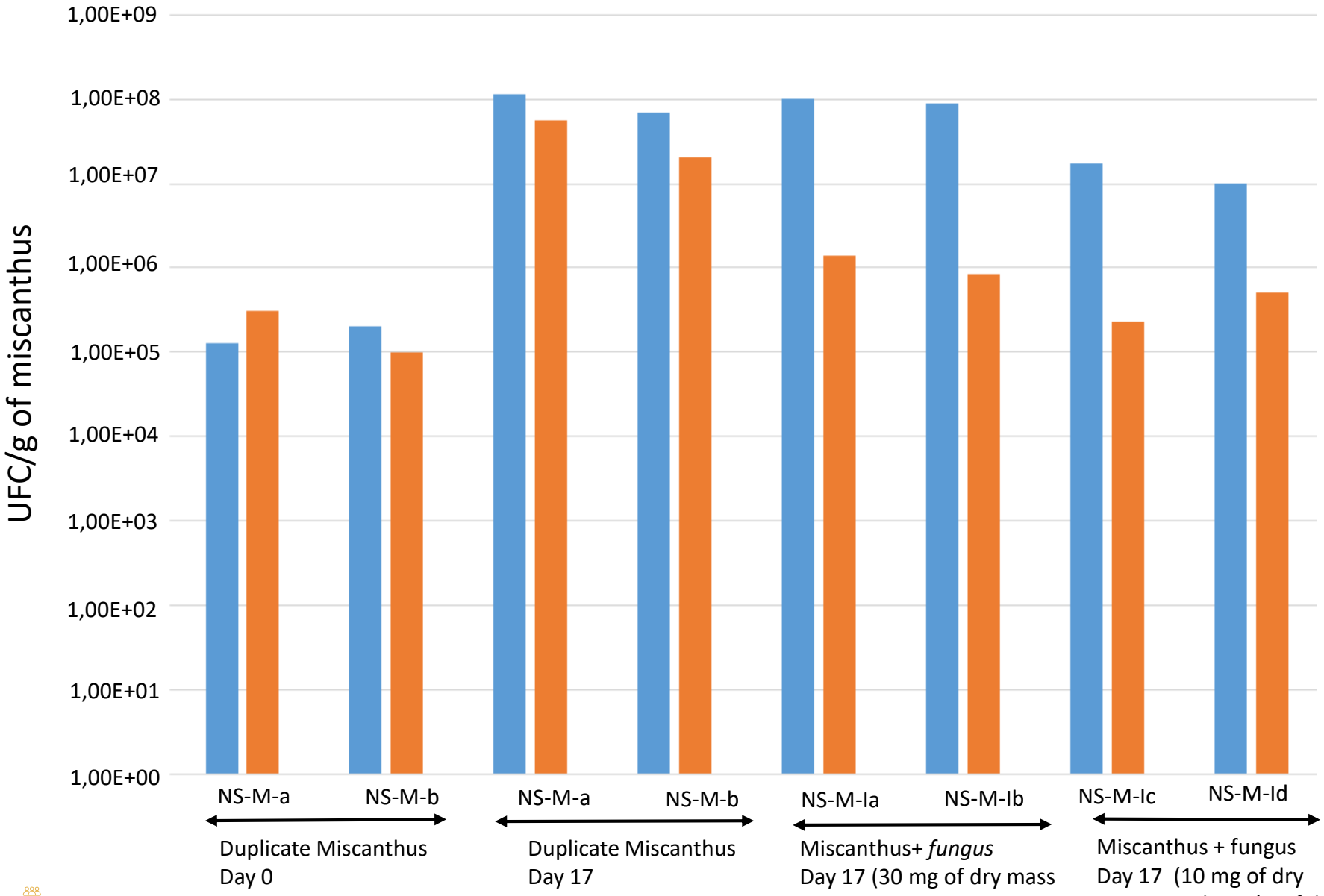
Evolution of the Fungal and Bacterial Population over Time in the Presence of the Fungus with different rate



Endogenous Bacterial Flora

Endogenous Fungal Flora

Non Sterilized Miscanthus :NS-M-a
Non Sterilized Miscanthus
Inoculated with the fugus:NS-M-I
25°C
90 % Relative humidity
17 days incubation

At 10 mg of dry mass mycelium / g of dry mass miscanthus, we observed the same effect as at 30 mg of dry mass mycelium / g of dry mass miscanthus

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Conclusion

Polyporus brumalis develops, even at a low inoculation rate, on non-sterilized miscanthus.

It has no effect on the bacterial flora, but it has a negative effect on the fungal flora, since it reduces its population.

2 hypotheses :

- *P. brumalis* is more competitive than the endogenous fungal flora for retrieving nutrients from miscanthus

- *P. brumalis* releases compounds that inhibit the growth of the fungal flora.

Perspectives

The production of the fungus inoculum is costly. It would therefore be important to determine the minimum inoculation rate, which would facilitate the implementation of the process and reduce its cost.

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Goals

AGRIMA aims to foster universities' **capacity building** for the **green transition** through **innovative practices and higher education curricula updating in agri-food waste management** for the **circular bioeconomy**.

AGRIMA addresses:

1. **Advancing pedagogical methods** for industrial agri-food waste valorisation **based on business-academia synergies**.
2. **Integrating citizen science** in bio-economy-enhanced waste valorisation as a means of **civic engagement and environmental advocacy**.

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