

## PRESERVATION OF FUNGI IN DISTILLED WATER PRELIMINARY RESULTS

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### SUMMARY

The Authors employed with success the CASTELLANI method of preservation of fungi in distilled water, for samples of *Paracoccidioides brasiliensis*, *Microsporium canis*, *Microsporium gypseum*, *Trichophyton rubrum*, *Trichophyton tonsurans*, *Trichophyton schoenleinii*, *Trichophyton concentricum*, *Trichophyton megnini*, *Nocardia brasiliensis*, *Sporotrichum schenckii*, *Cryptococcus neoformans*, *Streptomyces griseus*, *Epidermophyton floccosum*, *Histoplasma capsulatum*, *Phialophora pedrosoi*, *Candida albicans*, *Aspergillus niger*, *Penicillium notatum*, *Alternaria sp* and *Rhodotorula sp*.

The macro and micro characteristics of these fungi were studied during a period of 9 months, the longest so far observed by us, through periodical sub-culturing in agar-SABOURAUD, from samples maintained in distilled water. All the samples studied proved satisfactorily viable, especially the dermatophyta, which did not undergo any pleomorphism.

The number of samples studied was 24.

### INTRODUCTION

In 1939 CASTELLANI<sup>2</sup> obtained the first positive results on the preservation of fungi in distilled water, observing the good viability of the samples studied, belonging to the *Candida*, *Geotrichum*, *Epidermophyton*, *Cladosporium*, *Aleurisma* and *Actinomyces* genera. Later, the same author<sup>3,4</sup> found out that other fungi could also keep their viability when maintained in distilled water, the dermatophyta keeping their micromorphological characteristics and thus preventing the overcoming of pleomorphism, commonly occurring in this group of pathogenic fungi.

CASTAGNETTA & MUNGELLUZZI<sup>1</sup> verified that this was a very practical process for the preservation of fungi. Applying it to 98 different samples, these Authors observed the good viability of the organisms even after a 14 months period of maintenance in distilled water. CASTELLANI<sup>5</sup> turns to the subject again, calling the attention of microbiologists to this method of preservation of fungal strains and extending it to the preservation of enterobacteria, with good results.

BENEDEK (apud CASTELLANI<sup>5</sup>) confirmed the results obtained by this Italian researcher, referring that this process which allows

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subculturing at longer intervals affords advantageous results at least for the maintenance of fungal strains in small mycological collections.

The literature data available on these experiments of CASTELLANI are rare, much in contrast with the excellency of the method proposed by the Author. Of great plainness, CASTELLANI's process opens new views for the easy maintenance of fungal strains, mainly of dermatophyta, and possibly of other microorganisms.

#### MATERIAL AND METHODS

Twenty-four fungal samples (Table I) raised in agar-SABOURAUD at room temperature, were utilized. Some of them (dermatophyta) were of recent isolation.

TABLE I  
Preservation of fungal strains in distilled water

Fungi	Number of samples	Viability periods so far attained (months)
<i>Paracoccidioides brasiliensis</i>	5	9
<i>Microsporum canis</i> .....	1	8
<i>Microsporum gypseum</i> ....	1	9
<i>Trichophyton rubrum</i> .....	1	7
<i>Trichophyton tonsurans</i> ...	1	7
<i>Trichophyton schoenleinii</i> ..	1	7
<i>Trichophyton concentricum</i> .	1	5
<i>Trichophyton megnini</i> .....	1	3
<i>Nocardia brasiliensis</i> .....	1	7
<i>Sporotrichum schenckii</i> ....	1	7
<i>Cryptococcus neoformans</i> ..	1	6
<i>Streptomyces griseus</i> .....	1	3
<i>Epidermophyton floccosum</i> .	1	5
<i>Histoplasma capsulatum</i> ...	1	3
<i>Phialophora pedrosoi</i> .....	1	3
<i>Candida albicans</i> .....	1	5
<i>Aspergillus niger</i> .....	1	6
<i>Penicillium notatum</i> .....	1	5
<i>Alternaria sp</i> .....	1	6
<i>Rhodotorula sp</i> .....	1	6
Total .....	24	

Distilled water was poured into 5 ml cotton-stopped bottles and the whole sterilized by autoclaving (20 minutes, 120°C).

The seeding of these bottles was performed in sterile conditions; with the help of a platinum loop wire a small portion of the original culture was transferred to the distilled water bottle, care being taken to avoid the carrying over of the old culture medium. The bottles were stopped with rubber stoppers previously sterilized (20 minutes, 120°C in autoclave) and then sealed with aluminium cap sealing.

After labelling, these bottles were kept at room temperature. At one month intervals samples of these water preserved strains were transferred to agar-SABOURAUD. Time of growth, macroscopical and microscopical morphology of the resulting cultures were studied.

#### RESULTS

The results are assembled in Table I. All the samples submitted to the process kept their macro and microscopical characteristics and proved viable after intervals ranging from 3 to 9 months.

#### DISCUSSION

All the samples maintained in distilled water, some of them for 9 months, remained perfectly viable, being easily subcultured again in agar-SABOURAUD.

A fact that must be put in evidence is that the dermatophyta did not lose their original micromorphological characteristics, which assumes great importance if we consider how easily these organisms undergo pleomorphism when leading a saprophytic kind of life.

The results of our present experiments confirm that CASTELLANI's method, due to its great simplicity, should be better investigated mainly where fungi of industrial interest are concerned.

Small mycological collections can thus be easily maintained by this process even when poor material conditions are prevalent.

The behaviour of fungi submitted to the studied process of preservation, from a biochemical standpoint, as well as the extension of the method to other microorganisms, have not yet been investigated.

RESUMO

*Conservação de fungos em água destilada*  
*Resultados preliminares*

Utilizando o processo de CASTELLANI, da conservação de cogumelos em água destilada, observaram os AA. que amostras de *Paracoccidioides brasiliensis*, *Microsporum canis*, *Microsporum gypseum*, *Trichophyton rubrum*, *Trichophyton tonsurans*, *Trichophyton schoenleinii*, *Trichophyton concentricum*, *Trichophyton megnini*, *Nocardia brasiliensis*, *Sporotrichum schenckii*, *Cryptococcus neoformans*, *Streptomyces griseus*, *Epidermophyton floccosum*, *Histoplasma capsulatum*, *Phialophora pedrosoi*, *Candida albicans*, *Aspergillus niger*, *Penicillium notatum*, *Alternaria sp* e *Rhodotorula sp* preservam-se bem com esta técnica.

Examinando as características macro e microscópicas desses cogumelos, através de cultivos em ágar-SABOURAUD obtidos mensalmente a partir de água destilada, durante 9 meses, prazo máximo verificado até o presente momento, puderam observar viabilidade satisfatória de tôdas as amostras estudadas, principalmente em relação aos dermatófitos, que não sofreram pleomorfismo.

O número de amostras estudadas foi de 24.

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