

Culture of Glomales

Classical methodology

The most common method to propagate Glomales remains the pot culture technique. However, this methodology

- ➔ cannot guarantee contaminant-free material
 - ➔ is time and space consuming
 - ➔ requires regular assessment of viability and monospecificity of inoculum
 - ➔ complicates the acquisition of quality cultures
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Monoxenic culture

When grown on a suitable synthetic growth medium in association with excised roots, Glomales cultures can be maintained pure and viable for long term periods. Growth media commonly used are : the modified Strullu-Romand (MSR) medium (see Declerck *et al.*, 1998) which is a modification of the Strullu and Romand medium (1986) and the minimal (M) medium of Bécard and Fortin (1988).

Advantages of the monoxenic culture of Glomales

- ➔ **Reliability of cultures** : contaminant free cultures throughout life cycle
- ➔ **Monospecificity of fungal inoculum** : no cross contamination with other Glomales throughout life cycle
- ➔ **Non-destructive monitoring** : morphological observations conducted without disturbance of the intimate coexistence of host roots and AMF during whole life cycle
- ➔ **Efficient system for**
 - Physiological studies** : culture conditions are standardized thus facilitating studies on the impact of environmental factors
 - Molecular and biochemical studies** : optimal conditions due to the absence of undesirable contaminants
 - Mass-inoculum production** : feasible through regular subcultures
- ➔ **Lower space and time requirements** : produced under growth chamber controlled conditions
- ➔ **Easy quality control and fungal growth quantification** : important for high scale production of both *in vivo* and *in vitro* propagation

Inconvenients related to the monoxenic culture of Glomales compared to pot cultures

- ➔ **Sterile environment** : high precaution (controlled atmosphere, laminar flow hood, ...) are needed to succeed
- ➔ **Technical training** : may be necessary to acquire expertise
- ➔ **Low sporulation levels for some species** : make them less distributable or distributable at a prohibitive price
- ➔ **Number of available strains** : several strains are cultivated but few distributed
- ➔ **Continuous cultivation** : difficulty to maintain some strains under continuous growth
- ➔ **Physiological and genetic characters** : potential loss of strains characters due to successive transfers

Perspectives

With the availability for the scientific and the industry communities of high quality and contaminant-free AMF inocula, very innovative research and development possibilities, up to now neglected, will be greatly facilitated

- ➔ **By** much easier procedures for the maintenance of clean and reliable fungal culture
- ➔ **By** a much better control of inoculum quality and effectiveness
- ➔ **By** access to an experimental system that allows refined physiological and molecular investigations up to large-scale inoculum production

References

Bécard G. and Fortin J.A. (1988) Early events of vesicular-arbuscular mycorrhiza formation on Ri T-DNA transformed roots. *New Phytologist* 108 : 211-218.

Declerck S., Strullu D.G. and Plenchette C. (1998) Monoxenic culture of the intraradical forms of *Glomus* sp. isolated from a tropical ecosystem : a proposed methodology for germplasm collection. *Mycologia* 90 (4) : 579-585.

Strullu D.G. and Romand C. (1986) Méthode d'obtention d'endomycorhizes à vésicules et arbuscules en conditions axéniques. *C.R. Acad. Sc. Paris, t. 303, Série III. Vol. 6* : 245-250.

Glomales species cultivated monoxenically on root organ culture

Acaulosporaceae

**Acaulospora laevis* Gerd. & Trappe
A. morrowae Spain & Schenck
A. rehmi Sieverding & Toro

Declerck S (pers. comm.)
Wu C-G (pers. comm.)
Declerck S (pers. comm.)

Gigasporaceae

Gigaspora albida Schenck & Smith
G. gigantea (Nicolson & Gerd.) Gerd. & Trappe

G. margarita Becker & Hall

G. rosea Nicolson & Schenck
**Scutellospora castanea* Walker
S. nigra (Redhead) Walker & Sanders
S. reticulata (Koske, Miller, Walker) Walker & Sanders

Wu C-G (pers. comm.)
Gadkar *et al.*, 1997
Miller-Wideman and Watrud, 1984

Karandashov *et al.*, 1999
Forbes *et al.*, 1998
Declerck S (pers. comm.)
Wu C-G (pers. comm.)
De Souza (pers. comm.)

Glomaceae

Glomus aggregatum Schenck & Smith emend. Koske
G. caledonium (Nicolson & Gerd.) Trappe & Gerd.
G. cerebriforme McGee

G. clarum Nicolson & Schenck

G. constrictum Trappe

G. deserticola Trappe, Bloss & Menge
? *G. diaphanum* Morton & Walker

G. etunicatum Becker & Gerd.

G. fasciculatum (Thaxter sensu Gerd.) Gerd. & Trappe emend. Walker & Koske
**G. fistulosum* Skou & Jakobson

G. intraradices Schenck & Smith

G. lamellosum Dalpé, Koske & Tews
G. macrocarpum Tulasne & Tulasne

**G. mosseae* (Nicolson & Gerd.) Gerd. & Trappe

G. proliferum Dalpé & Declerck
G. versiforme (Karsten) Berch
Sclerocystis sinuosa (Gerd. & Bakshi) Almeida & Schenck

Moutoglis P (pers. comm.)
Karandashov *et al.*, 1999
Samson J, 2000
De Souza and Barbara, 1999

Wu C-G (pers. comm.)
Diop T (pers. comm.)

Mathur and Vyas, 1999
Mathur and Vyas, 1995
Declerck S
Schreiner and Koide, 1993

Pawlowska *et al.*, 1999

Karandashov *et al.*, 1999
Declerck *et al.*, 1998
Nuutila *et al.*, 1995

Gryndler *et al.*, 1998
Chabot *et al.*, 1992

Karandashov *et al.*, 1999
Dalpé Y (pers. comm.)
Declerck *et al.*, 1998
Mosse and Hepper, 1975

Douds, 1997
Declerck *et al.*, 1996b, 2000
Diop *et al.*, 1994
Wu C-G (pers. comm.)

+ non-identified *Glomus* sp. strains are maintained under root-organ culture

* non-sporulating species

? identification to be confirmed

