Hyphal tip growth and hyphal septation and Mycelium formation

Prepared by

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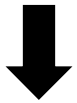
1. Protoplasm drown toward hyphal tip

- Hyphal growth polarization detection approaches:
  1. Electron microscopy.
  2. Video-enhanced microscopy of movement of green flourescent protein (GFP) tagged organelles in living hyphae.
  3. Fluorescence imaging of distribution of ions.
  4. Micro-electrode measurement of pH along hyphae.
  5. Patch-clamp detection of ion channels.
  6. Immunoflurescence detection of enzymes distribution.
  7. Measurement of turgor pressure by plasmolysis method.

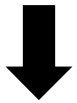
SpitzenkÖrper

- By phase contrast microscopy and vital staining of living hyphae with a membrane selective fluorescent dye

A cluster of small vesicles with no clear boundary was observed just beneath the plasma membrane of hyphal tip



This apical body is called SpitzenkÖrper
Collection center of vesicles containing enzymes & polysaccharides precursors



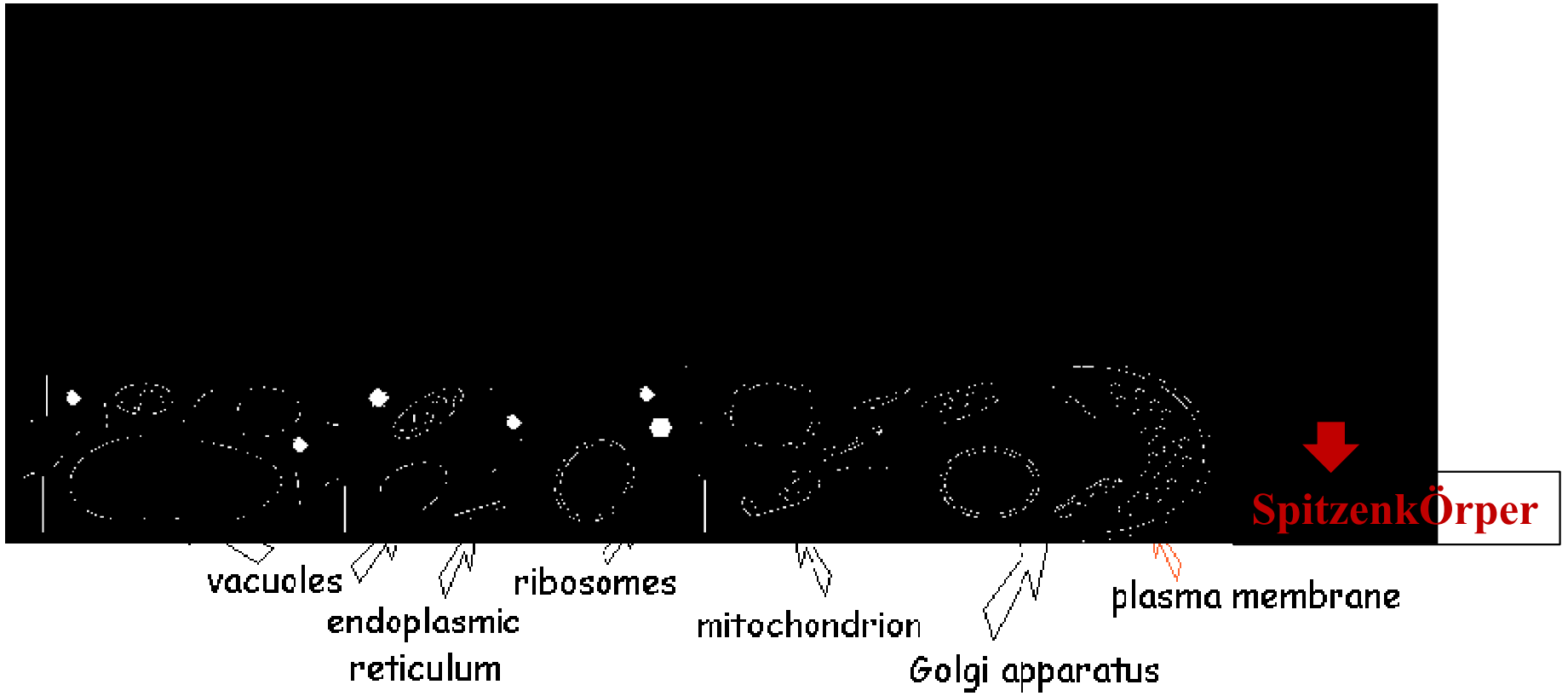
SpitzenkÖrper …. Advances continously as the hyphae elongates

Once SpitzenkÖrper discharge its contents …… new one

The cycle of collection and discharge of vesicles is consistent with the observation of hyphal growth as in pulses

Evidences :

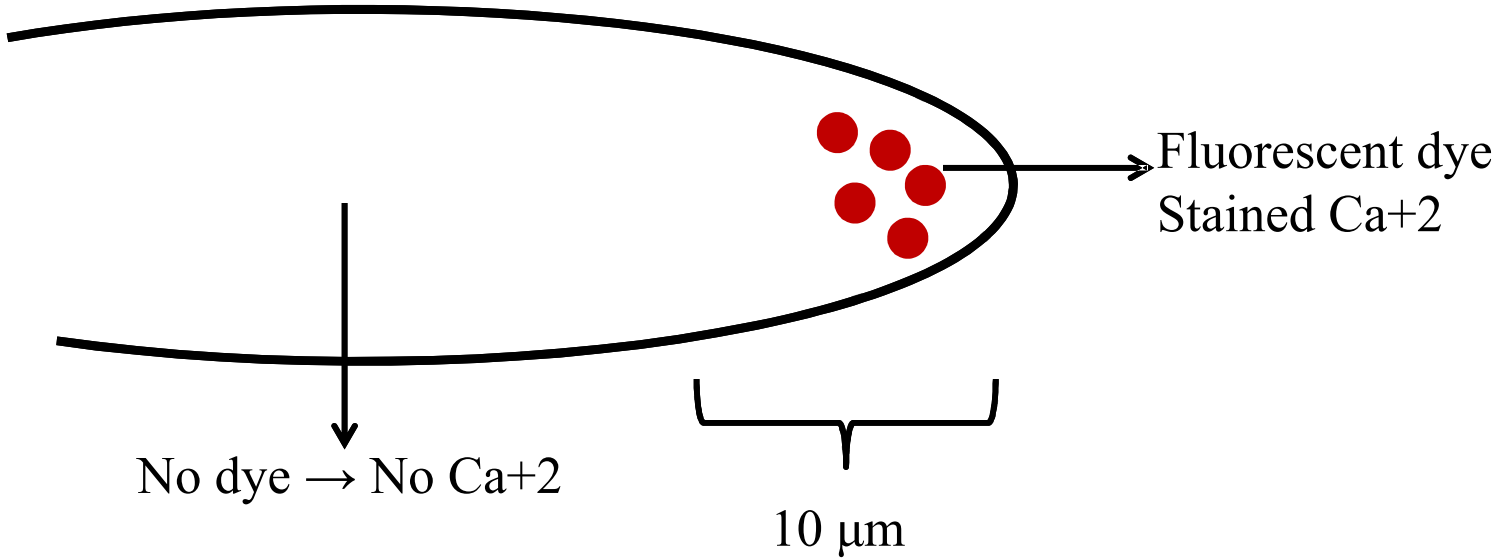
- **Video microscopy:**
  - Image analysis of hyphae showed close corelation of SpitzenkÖrper trajectory and the direction of hyphal growth.
  - SpitzenkÖrper association with meshwork of microtubules and microfilaments. So it is polarized trajectory determined by the growing scaffolding of microtubules.
- **Electron microscopy:**
  - Hyphal tip showed **Exocytosis** of Golgi-derived vesicles to growing tips which containing fungal wall synthesis required precursors allowing their delivery and polarized growth of hyphae.



Fungal tip organneles (www.fungionline.org.uk)

2. Tip-High Calcium

- As in other tip extending cells (plant root hairs, pollen tubes, rhizoid cells), fungal hyphal tip contain a tip-high gradient of **Calcium** ions.
- Evidences:
  1. The cytosolic [Ca+2] was measured by ratio imaging emission intensities of Ca+2-sensetive fluorescent dyes (fluo-3, fura red) by confocal microscopy.→ fluorescence emission was localized in the 10 µm region surrounding the tip of growing hyphae but not in the non-growing hyphae.



2. Hyphal elongation was inhibited by microinjection of Ca
   hyphal tip stoped elongation → tip hyphal gradient of free C
   for tip growth.

3. Mycelium formation

Hyphae of higher fungi extend via tip growth followed by cross-wall formation or septation whereas, lower fungi remain aseptate.

- **Septation.**
- **Branching.**
- **Hyphal fusion.**
- **Multihyphal structures**

1. Septation.

-fungi (except in Zygomycotina) hypha is partitioned into compartments by transverse walls = **septa** (**septum**).

- septa are spaced evenly.

- intercalary compartments have a uniform length of 38 µm→ no random septation.

- in fungi, the term compartment is often used in lieu of cells to denote that fungal cells have cytoplasmic continuity.

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1. Septa formation

-Septa are formed by **Centripetal growth** of fungal wall and have **Perforation**, through which cytoplasmic organelles, including nuclei can pass.

2. Septa function

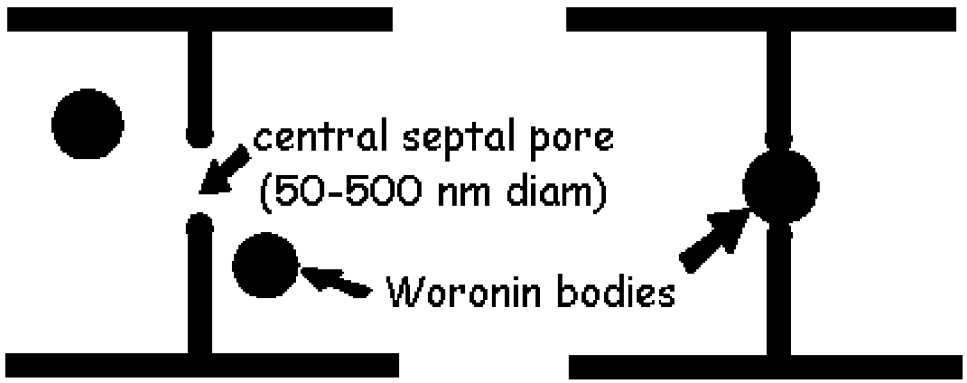
a- fungal hyphal structural support (as aseptated fungi e.g. zygomycetes & oomycetes are vulnerable to damage → contribution to hyphal rigidity.

b- enable differentiation (Septa can isolate adjacent compartments from one another so that different biochemical and physiological processes can occur within them - these may result in differentiation of the hyphae into specialized structures, such as those associated with sporulation).

c- first line of defense.

d-compartmentalize hyphae to reduce leakage when hyphae is ruptured or provide a reducing environment for redox-sensetive enzymes.

e- redirecting protoplasm movement to any region in mycelium by regulating closing or opening of septal pores by a **plug** of **Woronin Body** (perixisome derived dense core) on either septal pore [in case of subjection to damage or environmental stress] → → changing hyphal extension direction for productive exploration of nutrient-rich surroundings. → allow spatial regulation of branch sites and development of reproductive structures by redistribution of nutrients.



http://www.fungionline.org.uk/3hyphae/3septa.html

3. Septa types

Simple Septum

Relatively large
0.05-0.5 µm diameter

Allow passage of cytoplasmic organelles and even nuclei

Develop as ingrowing ring from the lateral walls of hypha which is associated with localized modification of lateral walls → localized proliferation of glycoprotein reticulum

Basidiomycetes & Ascomycetes
(Monokaryotic Hyphae)

Mutiperforate septum

9 nm diameter
/each micropore

These micropores allow cytoplasmic continuity between adjacent hyphal compartments, but are too small to allow cytoplasmic streaming to occur to the extent observed in fungi possessing larger septal pores.

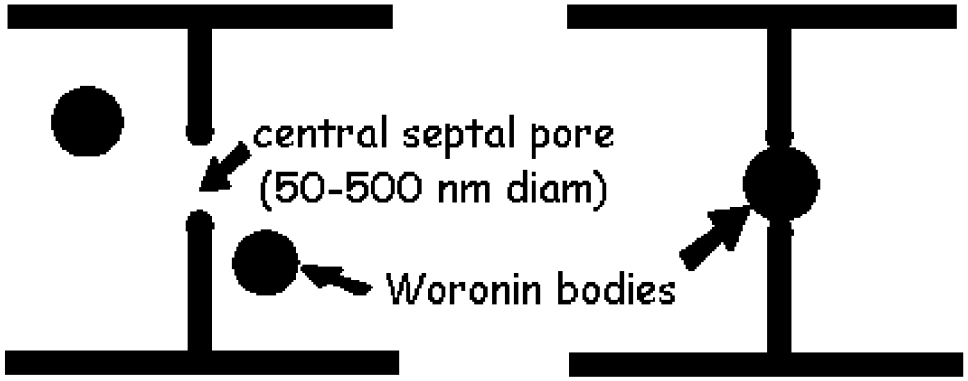
Dolipore Septum

Have narrow channel bounded by two flanges of amorphous glucan
100-150 nm diameter

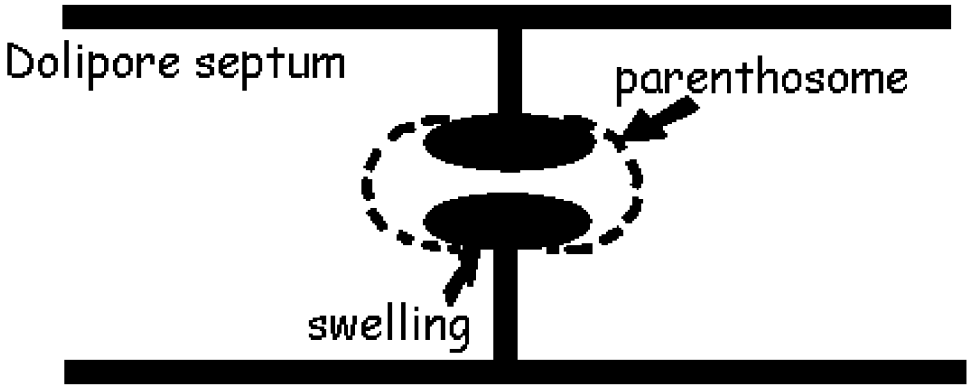
Allow cytoplasmic passage & continuity but prevent passage major organelles icluding nuclei

More complex as it is formed when strains of different mating compatibility groups fuse to form Dikaryon with two nuclei in each compartment.

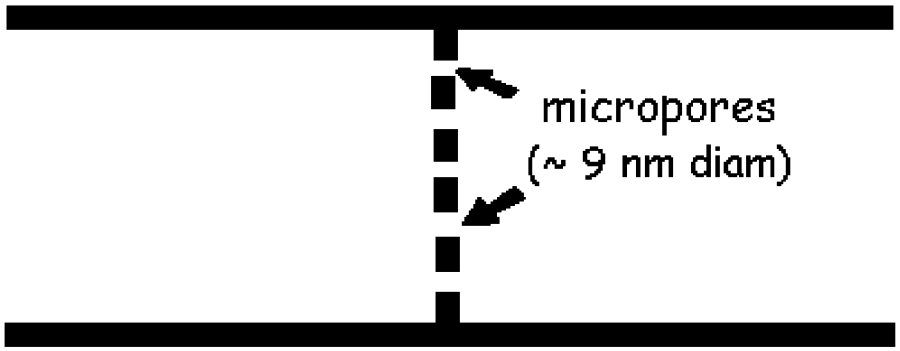
On ei…ped membr… parenthosomes



Simple Septum



Dolipore septum

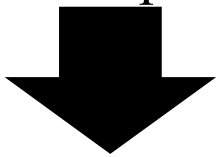


Multiperforate septum

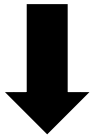
http://www.fungionline.org.uk/3hyphae/3septa.html

2. Branching

- Hyphal branches arise in acropetal succession in proximity → subapically to the septum.
- Hyphae tend to avoid their neighbors and grow outwardly from the center.
- Pattern of branching can be compared to the pattern of branching in a fir tree with a main hyphae and a series of branches borne alternately in two dimensions, suggesting a marked apical dominance.



As the leading hyphae diverge from one another



Apical dominancy becomes weaker

- Branching allow fungal colony to effectively colonizes and exploit its surroundings.
- As the original hyphae and the first order branches grow, produce further branches behind their tips → these branches divege from eventually colony develops a characteristic circular outline

Mechanism of branching

Evidence: genetic method

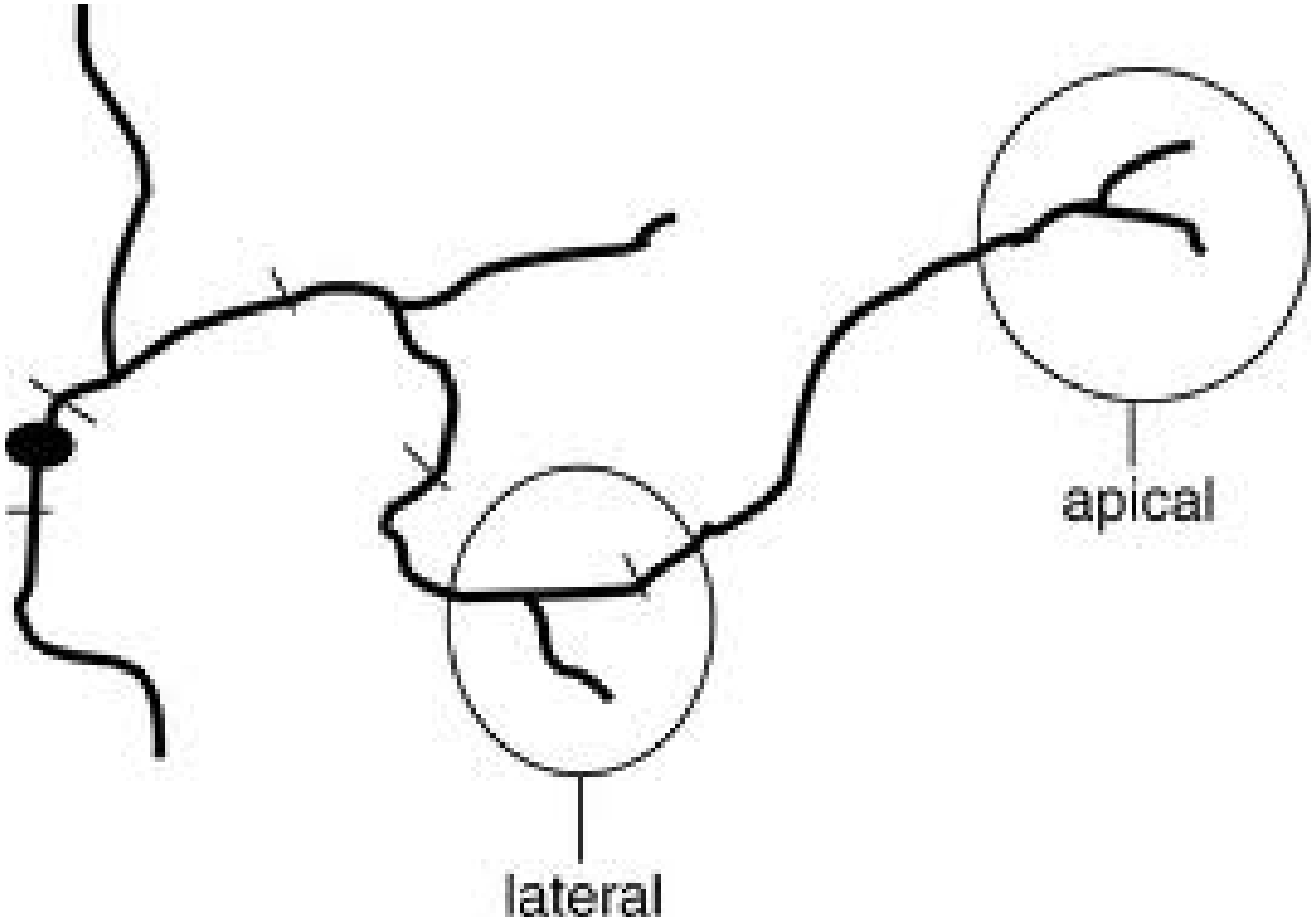
- Mutants → With defect in branching → Form compact colonies
- Wild types → Can branch → Spreading colonies

Involved in cellular organization of hyphar

Cytoplasm volume/nucleus is unaffected by substrate availability

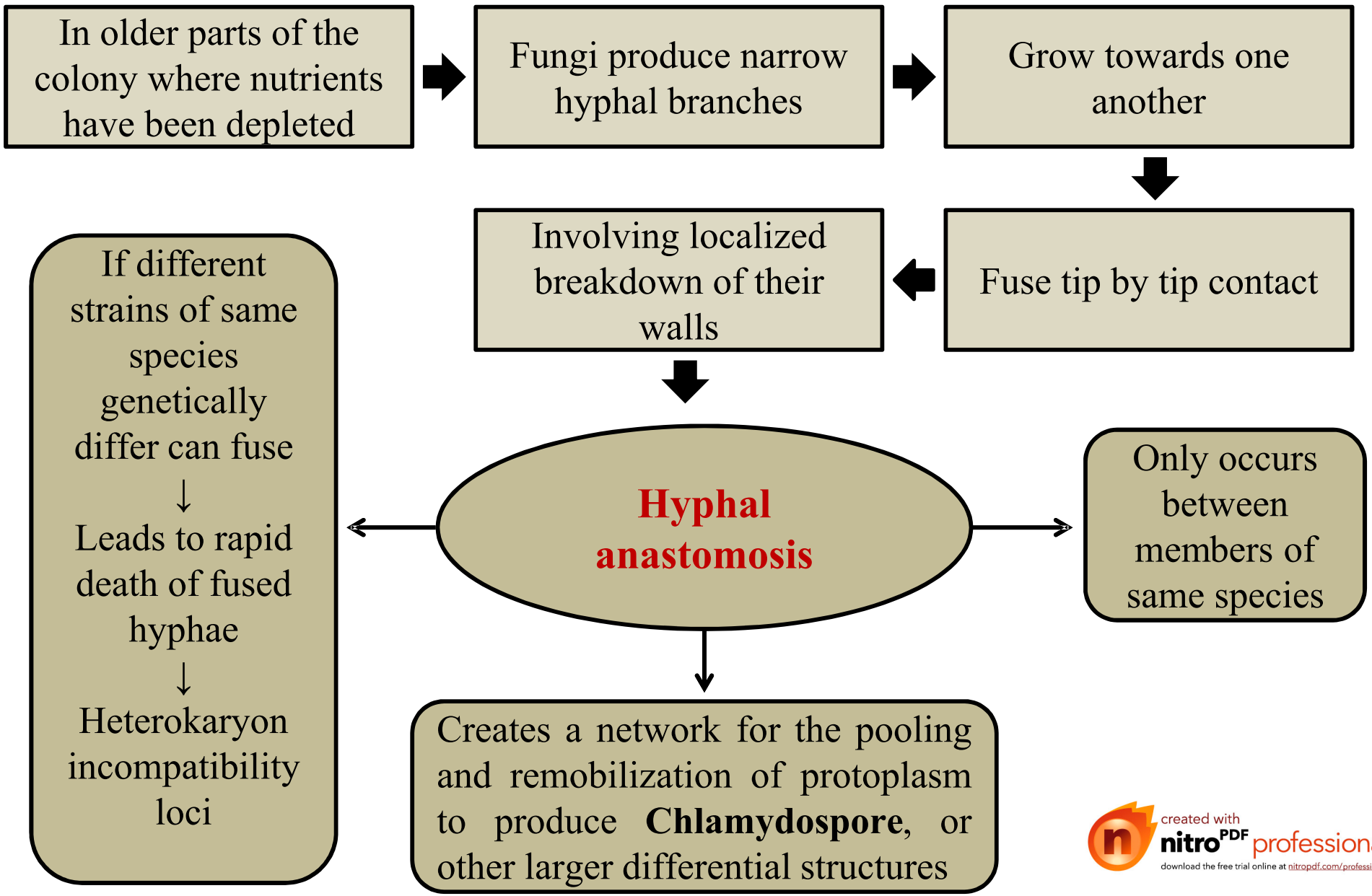
Bracnching and nuclear division are coordinated to maintain its ration

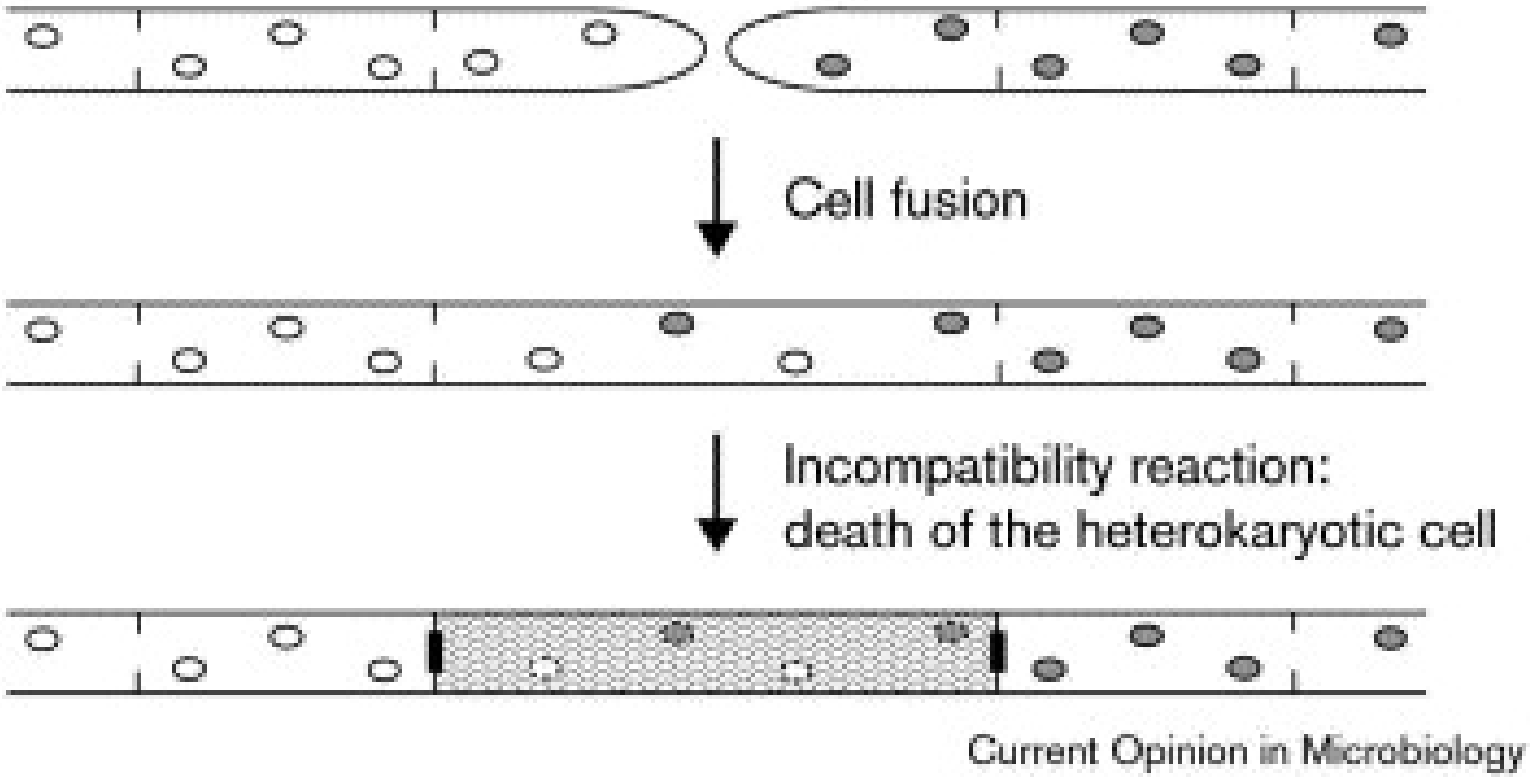
→ Nature of fungus can explore its surrounding with thin sparely branched hyphae → Minimum formation of Biomass



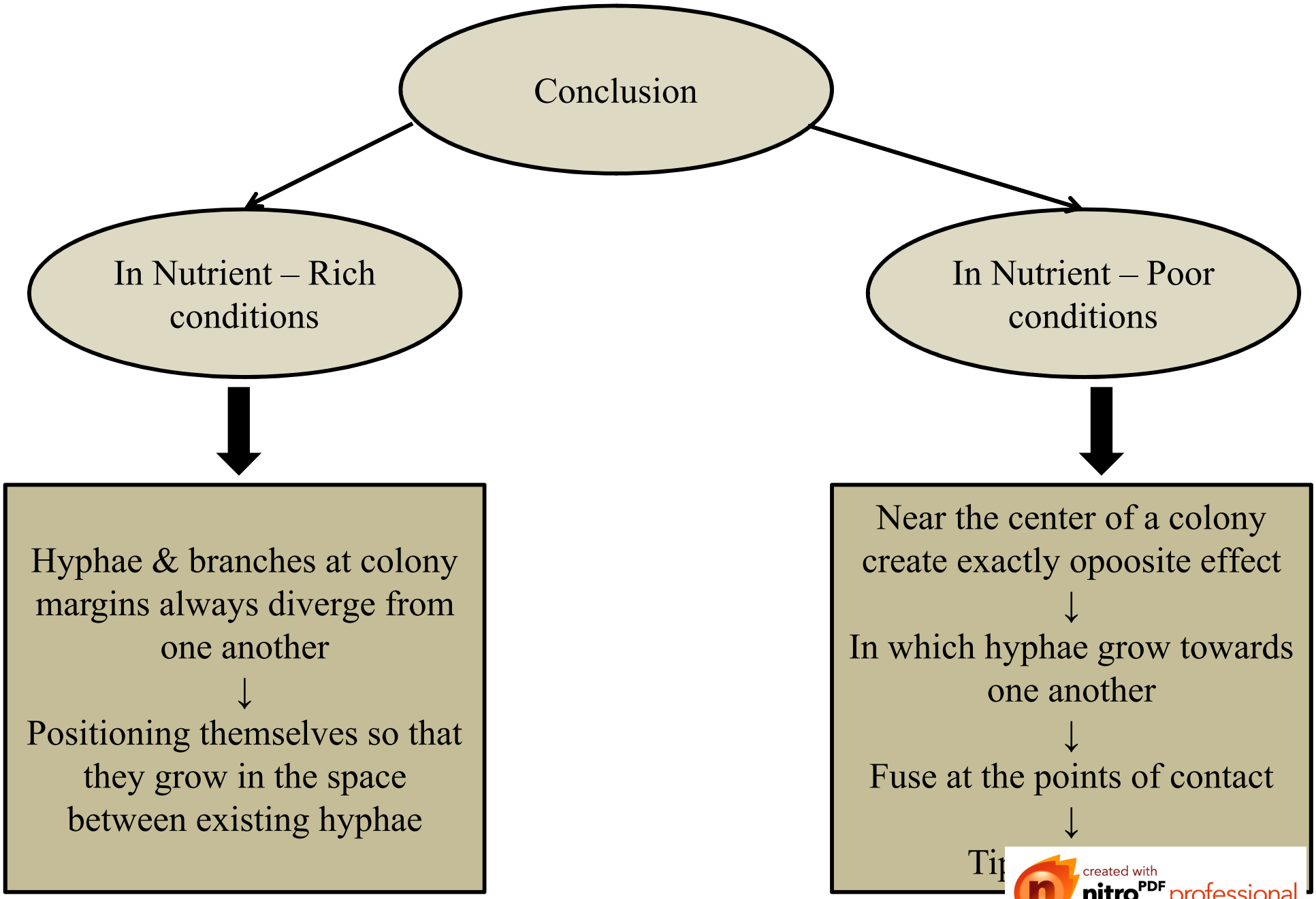
Fungal hyphal branching (www.mycologia.org)

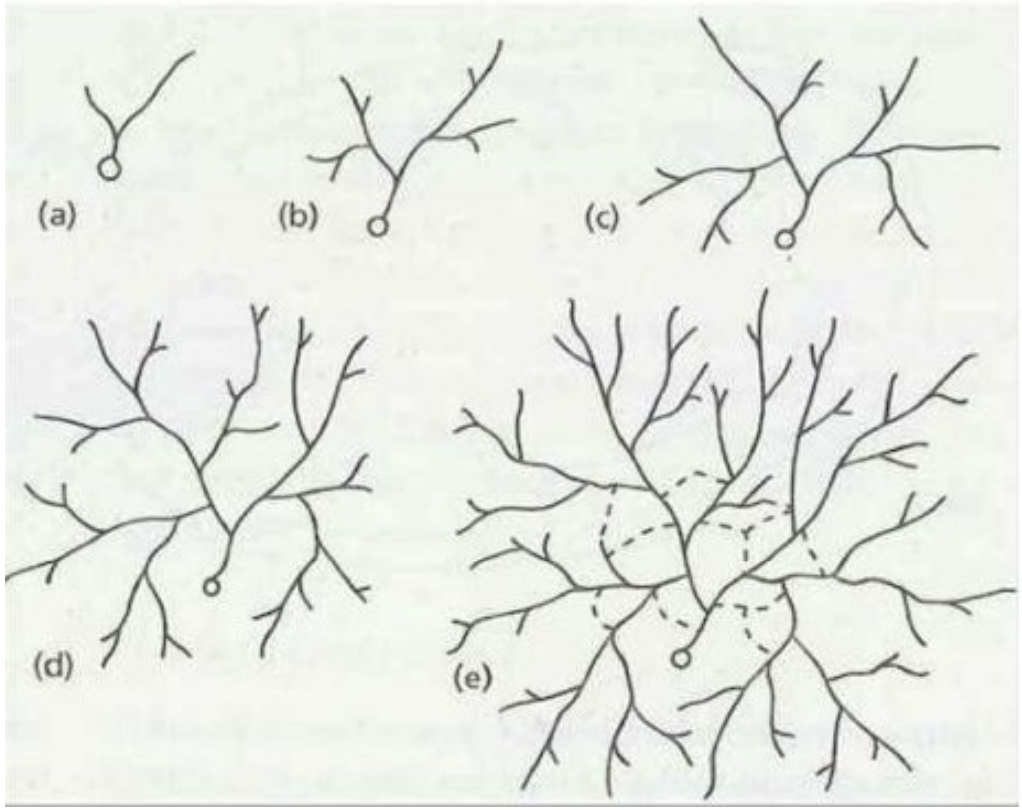
3. Hyphal fusion



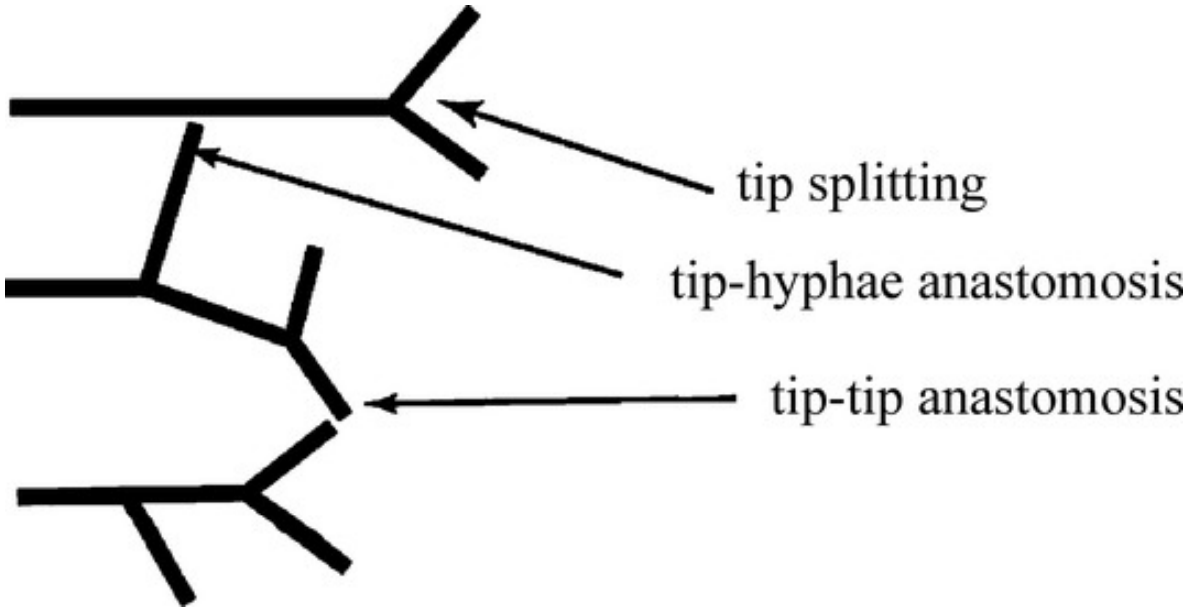


Fungal hyphal fusion (www.sciencedirect.com)



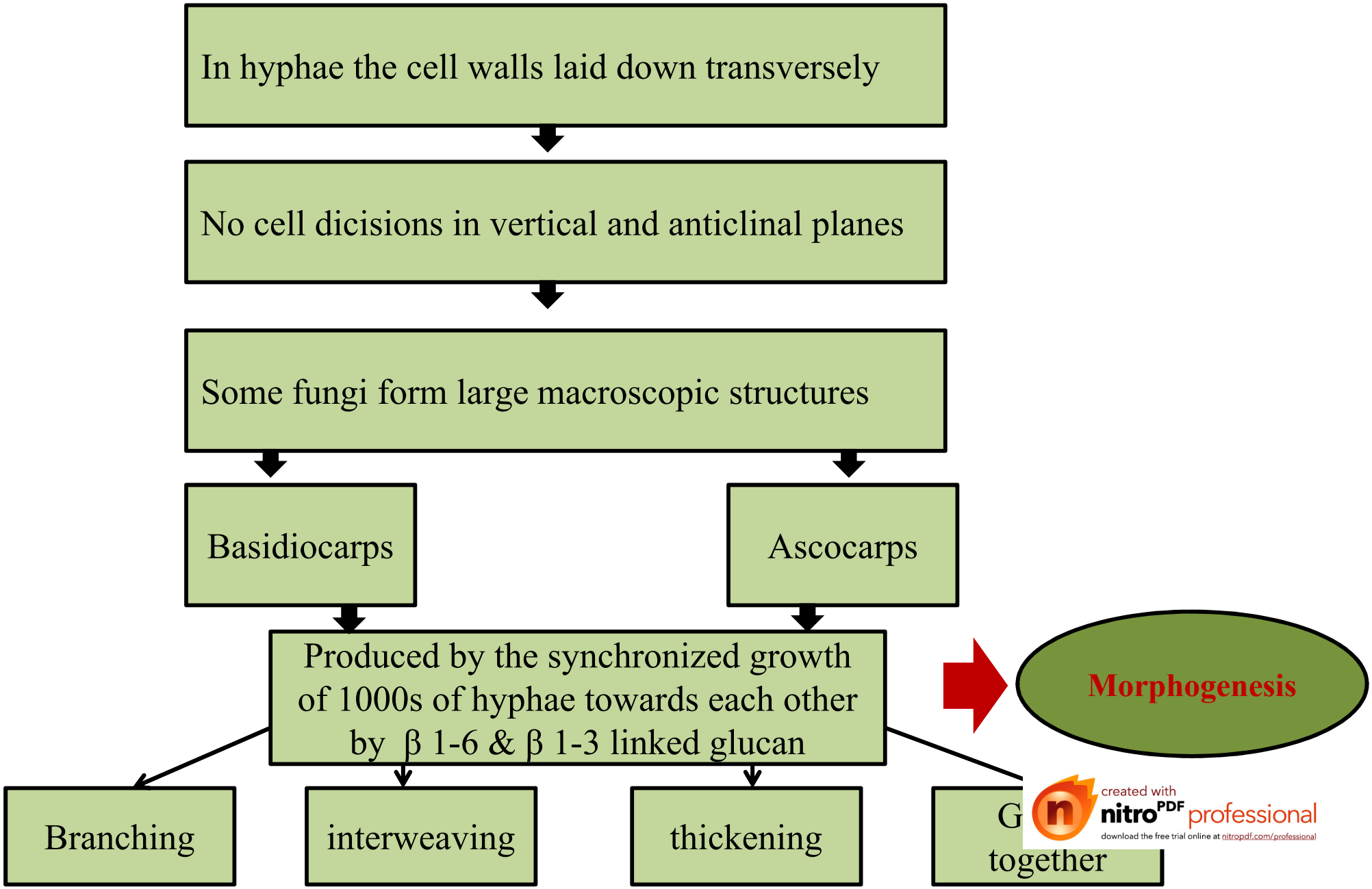


Fungal hyphal branching and anastomosus (www.bardoscalculus.com)



Schematic representation of the fungal growth processes, apical branching (tip splitting), tip–hyphae anastomosis and tip–tip anastomosis, included in the hyphal growth model of Schnepf et al. (2007) (www.springerimages.com)

4. Multihyphal structures



THANKS