Emerging multispecies fungal biofilms infection in clinical units Algeria

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Objectives : Our study aims to investigate the rate of mono or multi-species fungal alterations of medical devices implanted on patient presenting a risk of infection in neuro-surgery unit at hospital of Tlemcen Algeria. This study links the clinical setting with microscopic laboratory confirmation of the presence of dual species biofilm in carefully selected on the inner and outer surfaces of the peripheral venous catheters.

Materiel and methods

1. Samples and yeast isolates

Samples were taken from peripheral venous catheters (CVP) implanted for 48 hours or more. Isolated yeasts have been identified by Chromagar and API Candida (BioMérieux ®)

2. Biofilm s Formation

Mixtures of *C. albicans* and *C. glabrata* were prepared in the following ratios: 100/0 (*C. albicans* only) ; 90/10; 50/50; 10 : 90 and finally 0 : 100 (*C. glabrata* only) prepared in RPMI 640 / MOPS performed in 96-well polystyrene. Each experiment was done in triplicate. The protocol described by Ramage et al.(2001)

3.Reduction of tetrazolium salt :

The metabolic activity of *Candida* cells in biofilms was determined by the tetrazolium salt (XTT)

4. Optical Density Measurement (OD):

In accordance with the work of Valentin et al. (2007) and Villar-Vidal et al [2011], biofilm production is considered positive if the optical density measured at 492 nm ≥ 0.200 .

5. Observation by SEM of mixed biofilms:

The catheters are fixed by adding a solution of 2.5% (v / v) glutaraldehyde in PBS buffer. The samples are dehydrated in ethanol solutions 3x10 min at 30%, 50%, 70% and 100%. They are then brought to the critical point and then metalized with gold and observed under a scanning electron microscope HITACHI SU 3500

Fig 1 : dual species biofilm formed by *C. albicans* and *C. glabrata* with ratios of : 100:0; 90:10, 50:50, 1:90 & 0:100.

Results: This study revealed the mixed biofilm formation by dual species of *Candida sp.* co-isolated from the same catheter: *Candida albicans* and *Candida glabrata*. However, 2/3 of these evaluated dual-species biofilms are weak producers of biofilms.. The metabolic activity of dual- species biofilms increase whatever the proportion of the mixture from that found for single ones

Fig.2 : Segments of catheters metalized by gold for visualized by SEM (A) Micrographs taken by SEM of segment of catheter altered by mono and dual species *Candida* (B) and the structure of biofilms formed on catheters by *C. albicans* (C), *C. parapsilosis* (D), *C. glabrata* (E), *C. albicans* /*C. glabrata* biofilm formed on the external surface of a peripheral vascular catheter (F), the extracellular matrix (G), *C. albicans* /*C. glabrata* biofilm formed on the inner surface of a peripheral vascular catheter (H)

Conclusion: Dual species *Candida albicans* and *Candida glabrata* might be able to successfully co-exist on medical devices and generate mixed biofilms by developing synergistic or antagonistic relationships. This adds to the complexity of the treatments, sometimes very consistent