

## Effects of Culture Conditions on the Adhesion of Yeast *Candida* spp. and *Pichia* spp. to Stainless Steel with Different Polishing and Their Control

**Authors :** Ružica Tomičić, Zorica Tomičić, Peter Raspor

**Abstract :** An abundant growth of unwanted yeasts in food processing plants can lead to problems in quality and safety with significant financial losses. *Candida* and *Pichia* are the genera mainly involved in spoilage of products in the food and beverage industry. These contaminating microorganisms can form biofilms on food contact surfaces, being difficult to eradicate, increasing the probability of microbial survival and further dissemination during food processing. It is well known that biofilms are more resistant to antimicrobial agents compared to planktonic cells and this makes them difficult to eliminate. Among the strategies used to overcome resistance to antifungal drugs and preservatives, the use of natural substances such as plant extracts has shown particular promise, and many natural substances have been found to exhibit antifungal properties. This study aimed to investigate the impact of growth medium (Malt Extract broth (MEB) or Yeast Peptone Dextrose (YPD) broth) and temperatures (7°C, 37°C, 43°C for *Candida* strains and 7°C, 27°C, 32°C for *Pichia* strains) on the adhesion of *Candida* spp. and *Pichia* spp. to stainless steel (AISI 304) discs with different degrees of surface roughness ( $R_a = 25.20 - 961.9$  nm), a material commonly used in the food industry. We also evaluated the antifungal and antiadhesion activity of plant extracts such as *Humulus lupulus*, *Alpinia katsumadai* and *Evodia rutaecarpa* against *C. albicans*, *C. glabrata* and *P. membranifaciens* and investigated whether these plant extracts can interfere with biofilm formation. The adhesion was assessed by the crystal violet staining method, while the broth microdilution method CLSI M27-A3 was used to determine the minimum inhibitory concentration (MIC) of plant extracts. Our results indicated that the nutrient content of the medium significantly influenced the amount of adhered cells of the tested yeasts. The growth medium which resulted in a higher adhesion of *C. albicans* and *C. glabrata* was MEB, while for *C. parapsilosis* and *C. krusei* was YPD. In the case of *P. pijperi* and *P. membranifaciens*, YPD broth was more effective in promoting adhesion than MEB. Regarding the effect of temperature, *C. albicans* strain adhered to stainless steel surfaces in significantly higher level at a temperature of 43°C, while on the other hand *C. glabrata*, *C. parapsilosis* and *C. krusei* showed a different behavior with significantly higher adhesion at 37°C than at 7°C and 43°C. Further, the adherence ability of *Pichia* strains was highest at 27°C. Based on the MIC values, all plant extracts exerted significant antifungal effects with MIC values ranged from 100 to 400 µg/mL. It was observed that biofilm of *C. glabrata* were more resistance to plant extracts as compared to *C. albicans*. However, extracts of *A. katsumadai* and *E. rutaecarpa* promoted the growth and development of the preformed biofilm of *P. membranifaciens*. Thus, the knowledge of how these microorganisms adhere and which factors affect this phenomenon is of great importance in order to avoid their colonization on food contact surfaces.

**Keywords :** adhesion, *Candida* spp., *Pichia* spp., plant extracts

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