EFFECTS OF RADIATION AND SIMULATED MICROGRAVITY ON THE GROWTH OF CANDIDA ALBICANS

Obaid Ullah¹, Nellen Nwaobasi¹, Anu M. Mathew¹, John Ford², Robert Alaniz³, Miranda Hector¹, Shishir Shishodia¹, Jejelowo Olufisayo¹

¹Center for Bionanotechnology and Environmental Research (CBER), Texas Southern University, Houston, Texas, 77004, USA, ²Nuclear Engineering Department, 58A Zachry Engineering Center, Texas A & M University, College Station, Texas 77840 and ³Microbial and Molecular Pathogenesis, College of Medicine, 417B Joe H. Reynolds Medical Bldg, Texas A&M Health Science Center, College Station, Texas 77843-1114.

ABSTRACT

We report preliminary data on the growth of Candida albicans following simultaneous exposure to both radiation and microgravity. The C. albicans cells (1x10⁶/ml) were cultured in Sabouraud Dextrose Broth. Growth was examined every 24 hours over a 4 day period using optical density and viability assays. Each experimental group received different exposures to both radiation and microgravity. Group 1 (Control 1g) cultures were not exposed to microgravity or radiation. Group 2 (SMG) cultures were exposed to simulated microgravity inside a Slow Turning Lateral Vessel (STLV), but not to radiation. Group 3, (radiation only) cultures were exposed to 25 mRad of gamma radiation per hour over a period of time to attain a range of radiation doses as follows: 24h = 0.6Rad; 48h = 1.2Rad; 72h = 1.8Rad; 96h = 2.4Rad. Group 4 (SSF) cultures received simultaneous exposure to microgravity and gamma radiation over a 4 day period as follows: 24h = 1.02Rad; 48h = 2.04; 72h = 3.06 Rad; 96h = 4.08 Rad. Group 5 (ISSF) received simultaneous and intermittent exposure to microgravity with gamma radiation exposure as follows: 24h = 1.02Rad; 48h = 1.02; 72h = 2.04 Rad; 96h = 3.06 Rad. SMG caused an increase in both biomass and optical density. Flow cytometry was also used to identify metabolically active cells; metabolically inactive cells; unstained cells; as well as cells that picked up both metabolically active and inactive cells. For cells in the control group, metabolic activity increased from 0 at inoculation, to 37.45 %, one day after inoculation; to 69.6% 2 days after inoculation and 93.05% 3 days after inoculation. Metabolic activity was higher under SSF (55.7%) than control by one day after inoculation, increasing to 67.8% by day 2 and 89.7% by day 3. Metabolic activity was highest under ISSF (61.05%) one day after inoculation, increasing to 67.05% by day 2 and 87.4% by day 3. Thus SSF and ISSF caused increased metabolic activity especially during the first 24 hours of exposure resulting in the observed shorter lag phase and increased growth. Formation of pseudo hyphae, bud formation and clumping were observed in all samples. However, the percentage of cells forming pseudohyphae or germ tubes after 96 hrs was 1.78% in control; 23 % in SMG; 2.33 % in gamma radiated cells; 22% in SSF and 30% in ISSF. Clumping was more prevalent within SMG and SSF while formation of germ tubes and pseudophyphae was observed more frequently within ISSF. Simulated microgravity alone and when applied simultaneously with gamma radiation resulted in increases in biomass yield and optical density measurements. C. albicans formed pseudohyphae, germ tubes, and showed greater tendency towards filamentous growth when grown under SMG, ISSF or SSF. Gamma radiation alone did not cause significant change in biomass yield to optical density when compared to the control. The radiation only experiment actually received a lower dose than SSF and ISSF and this may be a factor in the observed differences in growth and morphology. Morphological Changes observed for SMG, SSF and ISSF cultures include alterations in budding pattern, cell clumping, and increased frequency of hyphae and pseudohyphae. Cell clumping sometimes results from defects in mother-daughter cell separation. Viability assay based on metabolic activity measurements using the flow cytometer was higher in SSF and ISSF one day after inoculation, the SSF and ISSF environments might have stimulated early cellular responses for survival. However, metabolic activity for all groups continued to increase until day 4 when they reached their limits and the control had a higher percentage of metabolic activity. The mean population of dead cells in the SSF and ISSF were higher than that of the control sample but the change was not significantly different. The C. albicans cells under SSF and ISSF were clumped together compared to the control and yielded higher biomass as well as higher optical density readings. Under SSF and ISSF cells increased formations of germ tubes, hyphae, and pseudohyphae. The data suggests that simulated spaceflight induces alterations to yeast cell function that results in phenotypical alterations of growth and morphology.