

Post cryopreservation screening of sorghum microspores on MS media with various antibiotics

Tissue culture is sensitive to infectious micro-organisms through multiple avenues including improper sample handling, non-hygienic workspaces, and cross contamination. Sorghum tissue culture is hampered by contamination and antibiotics are expected to reduce bacterial issues for cryopreserved microspores. Using antibiotics can prevent the growth of bacteria and fungi which minimizes the loss of cell cultures. Sterile MS media 5501 in 200ml sections were mixed with each of the four antibiotics tested for efficiencies. Aliquots of 20ul (replicates) recovered concentrated microspores were placed on each plate which were observed at least for 24 hours to assess each treatment. Sorghum microspores were extracted in MS media using mortar and pestle and spun down to concentrate density. Even under sterile conditions, when plated, microspores can display signs of contamination. Four antibiotics (Carbenicillin, Cefotaxime, Hygromycin B, Kanamycin) and one plant protective mixture (anti-fungal) were screened towards the reduction of bacterial and fungi growth. The plant protective mixture used as treatment reduced fungal invasions in over 90% for all plates tested. The four antibiotics tested showed various success, with none of those eradicating the bacterial infections completely. Microspore growths were sub-cultured frequently on fresh plates which ultimately cleared infection, however, this, practice reduced the capacity of cells for selection dramatically.