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PURIFICATION OF TOXINS FROM BACTERIAL ISOLATES OF FRUIT AND VEGETABLE SALADS

The study aimed at the purification of toxins produced by isolates from street fruit and vegetable salad samples. Four cultures were selected, out of 33 cultures, based on protein analysis, hemolysis, and antibiotic susceptibility tests. The selected cultures were identified based on morphological and molecular analysis. The isolate SS07-4 as *Pseudomonas aeruginosa*, the isolate SS08-1 as B*acillus licheniformis*, the isolate SS01-6 as *Mammaliococcus sciuri*, and the isolate SS03-5 as *Escherichia coli.* Further, PCR analysis was carried out on each isolate to check the presence of specific virulence genes. Based on the PCR results, the *Pseudomonas aeruginosa* isolate confirmed positive for the phzS, algD, toxA, exoY, and exoT genes, and the *Bacillus licheniformis* isolate was confirmed to be positive for the LicA, LicB2, and LicC genes, which were considered for purification of their specific toxin. The purification of proteins from *Pseudomonas aeruginosa* and *Bacillus licheniformis* were carried out using a sequential approach involving ammonium sulphate precipitation, dialysis, gel filtration, and ion exchange chromatography. The concentration of protein from *Pseudomonas aeruginosa* in crude sample was found to be 5.1mg/ml, ammonium sulphate precipitated sample was 0.20 mg/ml; dialysed sample was 0.48 mg/ml, ion exchange chromatography sample was 0.45 mg/ml, and gel filtration sample was 0.04 mg/ml. Whereas the concentration of protein from *Bacillus licheniformis* in crude sample was 4.2 mg/ml, ammonium sulphate precipitated sample was 0.25 mg/ml, dialysed sample was 0.58 mg/ml, ion exchange chromatography sample was 0.36 mg/ml and gel filtration sample was 0.39 mg/ml. The crude and purified toxins were evaluated for their protein profile using SDS-PAGE to check the effectiveness of the process.

Authors: AMARANATH, Chinmayi (JAIN(Deemed -to-be University)); Ms M.S., USHA (JAIN(Deemed -to-be University))

Presenter: AMARANATH, Chinmayi (JAIN(Deemed -to-be University))

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