



ABSTRACT

The present study discussed the production and characterization of an extracellular laterosporulin (LS) from a native bacterial isolate inhabited in acidophilic soil. Soil samples were collected from four different habitats i.e. garden area, irrigation area, industrial area and hilly area of Murree, Punjab, Pakistan for isolation of bacteria. After primary and secondary screening, ISL-4 (0.53 ± 0.03 IBU/ml) and ISL-7 (0.60 ± 0.03 IBU/ml) were selected having better potential for LS production. Bacterial isolates ISL-4 and ISL-7 were identified to be *B. laterosporus* by biochemical test and electron microscopy. Various physical and cultural parameters were optimized to enhance the production of LS by *B. laterosporus*. A significant ($p \leq 0.05$) increase in LS production was observed when the physical parameters including medium volume, initial pH (7.0), size of inoculum (2 ml), incubation time (48 h), buffer pH (7.0) and incubation temperature (37°C) were further optimized. The effect of stabilizing agents (tween 20, tween 80, Na-EDTA, sodium dodecyl sulfate, 2-mercaptoethanol, triton X-100, glycine, vitamin B complex) for evaluation of LS activity was observed. The activity was maximum when treated with glycine where it was 7.92 ± 0.39 and 8.45 ± 0.42 IBU/ml for ISL-4 and ISL-7, respectively. The role of organic solvents (ethanol, methanol, butanol, propanol, chloroform, n-hexane, acetone, carbon tetrachloride) was also determined. The addition of 0.5 ml chloroform enhanced the activity to 10.26 ± 0.51 for ISL-4 and 11.31 ± 0.56 IBU/ml for ISL-7 which is highly significant (*HS*, $p \leq 0.05$). An overall enhancement of 18.83 fold was observed after optimizations of cultural conditions and additions of various stabilizing agents. Mass spectrometry analysis of LS was also done that was confirmed to be 5.605kDa. Antimicrobial potential of LS was also exploited using various techniques. Thus it can be concluded that *Brevibacillus sp.* isolated from acidophilic soil possess strong potential for LS production.