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LC-MS/MS analysis

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1 Works for me dx.doi.org/10.17504/protocols.io.bjdvki66

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LC-MS/MS analysis

Nano-LCMS using 6540 UHD Accurate Mass QTOF (CAMS, Venture Center, NCL Pune, India) was used to carry out all our mass spectrometric analysis. The peptides were analyzed using reversed-phase nanoscale liquid chromatography coupled to tandem mass spectrometry. The system consisted of a column; initially, the sample would be subjected to Agilent Binary LC1290 using column Polaris, high-performance chip, 360 nl enrichment chip (150mm X 75µm), and a separation column G4240-62030 with a thermostat temperature of 4°C. The mobile phase was Water (0.1% Formic Acid) and Acetonitrile: Water (90:10 v/v). The chromatography was carried out in a gradient manner (3% to 90% Acetonitrile: Water). The mass spec analysis was carried out on Agilent 6540 UHD QTOF MS (Gas temp: 250, Gas flow (l/minute): 8 minutes,) MS analysis was carried out in a data-dependent manner with survey scan resolution. Mass range: 250-1700(m/z), MS/MS: 50-1700(m/z), Data type: Centroid and software: Masshunter workstation software. Obtained data were searched against the UniProt protein databases of all the respective four Bacillus sp. using default parameters.