M Faisal Shahid et.al., J Mol Genet Med 2018, Volume 12
DOI: 10.4172/1747-0862-C1-025

Minimal step approach for phosphate free purification of FtsZ GTPase from *Salmonella typhimurium* and its functional characterization and optimization of rapid screening GTPase assay

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Bacterial cell division is pivotally regulated by a tubulin homolog protein FtsZ. This protein is a GTPase and polymerizes at cell center after nucleoid division to evenly divide the growing cell. Inhibition of FtsZ GTPase activity directly halts bacterial division. Due to this property, FtsZ has been validated as a drug target. However, purification of FtsZ requires dialysis step to remove residual phosphate from its binding pocket which cause overall protein loss. GTPase assays available also rely over specially purified GTP substrate and kit based methods. A minimal step approach from cloning, expression, purification, phosphate removal and GTPase assay establishment is presented for *Salmonella typhimurium* FtsZ protein. The assay has been established with trans-cinnamaldehyde, a natural compound with an IC50 value of 7.8 μM in 96-well format. The method circumvents the need of dialysis, and does not require kit based reagents for GTPase activity evaluation. Moreover, the assay has been optimized for with different concentrations of dimethyl-sulfoxide (DMSO), ethanol and methanol which can be used as solvents in high throughput compound screening for inhibition-based studies in anti-microbial development research projects.

Biography

M Faisal Shahid is currently pursuing his M Phil in Molecular Medicine from PCMD, ICCBS. He is also working as a Lecturer at the Department of Biosciences, Mohammad Ali Jinnah University, Pakistan. He has worked on molecular diagnosis of viral hepatitis and Mycobacterium tuberculosis.

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