

XIX CONGRESO DE LA SOCIEDAD ARGENTINA DE MICROBIOLOGÍA GENERAL

22 al 25 de octubre del 2024

Centro cultural y Pabellón Argentina de la Universidad Nacional de Córdoba, Córdoba, ARGENTINA.



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OPTIMIZED DNA EXTRACTION PROTOCOL FOR *Staphylococcus aureus* USING LIQUID NITROGEN

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Efficient and cost-effective DNA extraction is crucial for high-throughput experiments like whole-genome sequencing. *Staphylococcus aureus*, a gram-positive bacterium with a thick peptidoglycan layer, poses a challenge due to its resistance to enzymatic lysis. Traditional methods often rely on lysostaphin, an effective but expensive enzyme. This study presents an optimized DNA extraction protocol using liquid nitrogen for the lysis of *S. aureus* cells, including the USA 300 reference strain and clinical isolates from children with cystic fibrosis. The protocol involves cold mortarization with liquid nitrogen, followed by a phenol-chloroform extraction process. This method yielded high-quality DNA with average concentrations of $1413.2 \pm 553,8$ ng/ul, meeting all purity and integrity criteria necessary for molecular biology assays. The DNA integrity was confirmed via agarose electrophoresis, and purity was validated using spectrophotometric measurements, with 260/280 and 260/230 ratios indicating minimal contamination. This protocol offers a cost-effective alternative to enzymatic lysis, providing a reliable method for obtaining pure and concentrated DNA suitable for downstream applications.

Palabras clave: DNA extraction-*Staphylococcus aureus*-liquid nitrogen-phenol-chloroform-bacterial lysis