

Resource Summary Report

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RNA isolation and cDNA synthesis from the marine ichthyosporean *Sphaeroforma arctica*

DOI:10.17504/protocols.io.wqdfds6

Type: Protocol

Proper Citation

Sebastian Najle 2019. RNA isolation and cDNA synthesis from the marine ichthyosporean *Sphaeroforma arctica*. protocols.io dx.doi.org/10.17504/protocols.io.wqdfds6

Protocol Information

URL: <https://dx.doi.org/10.17504/protocols.io.wqdfds6>

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Summary: This protocol describes the steps for isolation of total RNA for cDNA synthesis in the marine Ichthyosporean *Sphaeroforma arctica*. In general, Ichthyosporeans are characterized by having a thick cell wall, of unknown composition, that is particularly hard to break, making cell lysis difficult. Here we have introduced a simple modification on the conventional RNA extraction protocol using TRIzol reagent, by including two cycles of freezing-thawing of the cells-TRIzol suspension, in order to facilitate cell lysis. This modification improved significantly the RNA isolation yield. Total RNA is further treated with DNase I enzyme and purified by precipitation to minimize genomic DNA contamination. Highly pure total RNA is then used to synthesize cDNA for downstream applications.

Associated Publications: Ocaña-Pallarès E, Najle SR, Scazzocchio C, Ruiz-Trillo I (2019) Reticulate evolution in eukaryotes: Origin and evolution of the nitrate assimilation pathway. PLoS Genet 15(2): e1007986. doi: [10.1371/journal.pgen.1007986](https://doi.org/10.1371/journal.pgen.1007986)

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