

Product datasheet

Hypotonic and Lysis Buffer for sonication

Catalogue no: 100004/100008

Applications: ChIP, Functional studies

Storage: +4°C

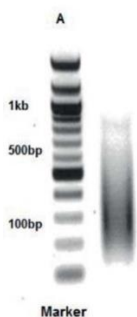
Size: 10/50 ml

Buffer preparation: Lysis Buffer for sonication must be pre-warmed to 40°C in a water bath for 30 minutes with occasional agitation before use, to remove any precipitates. The contents of the bottle should be mixed by inverting, bring the buffer back to room temperature when ready to use. The volume of buffer required for chromatin preparation is dependent upon the application and starting cell number.

General notes:

Chromatrap®'s Hypotonic and Lysis Buffer for sonication ensures the complete disruption of the nuclear and cell membrane of both adherent and suspension cultured mammalian cells required for chromatin immunoprecipitation (ChIP). The Lysis Buffer contains 1.0% wt/vol SDS, optimal for the sonication of chromatin samples. To determine how much Hypotonic and Lysis buffer for sonication to use refer to the manual <https://www.chromatrap.com/support/download/2016-06-06-25-1-chromatrap-spin-column-chip-ki.pdf/>

To prevent antibody inhibition the volume of Lysis buffer should not exceed 10% of the total slurry volume in the IP step.



Hec50 chromatin extraction using Chromatrap® Hypotonic and Lysis Buffer for sonication 100004.

5 million Hec50 cells were grown to ~80% confluency before cross-linking in 1% formaldehyde, quenching with glycine and collection in ice-cold PBS. Cells were spun down and the supernatant discarded before re-suspension and cell membrane lysis in 800µl Hypotonic Buffer to release the nuclei. Nuclei were lysed using 300µl of Lysis Buffer and the cell debris separated by centrifugation. The chromatin was sheared using a water bath sonicator with 30 second bursts with 30 second intervals at a power setting of 3 for 15 minutes to achieve optimal fragment lengths of 100-500bp.

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