Chelex DNA extraction Protocol

Sarah Chang[1], Michael Russello[1]

[1]University of British Columbia

Dec 24, 2021

1

dx.doi.org/10.17504/protocols.io.byvhpw36

Sarah Chang

Laboratory protocol for DNA extraction from animal tissue with Chelex.

DOI

dx.doi.org/10.17504/protocols.io.byvhpw36

https://doi.org/10.1371/journal.pone.0261966

Sarah Chang, Michael Russello 2021. Chelex DNA extraction Protocol. **protocols.io** https://dx.doi.org/10.17504/protocols.io.byvhpw36

protocol

Chang SL, Ward HGM, Russello MA (2021) Genotyping-in-Thousands by sequencing panel development and application to inform kokanee salmon (*Oncorhynchus nerka*) fisheries management at multiple scales. PLoS ONE 16(12): e0261966. doi: 10.1371/journal.pone.0261966

protocol ,

Oct 07, 2021

Dec 24, 2021

53897

1 Make up enough 10% Chelex for entire plate (**195 µL** /individual + slop) (ie. **1 g** Chelex: **9 mL** dH2O)

2 Place a magnetic stir bar in the jar containing your 10% Chelex solution. The Chelex powder is very heavy and needs to be constantly stirred in order to maintain consistency.

3 Pipette **195 µL** of Chelex into each well of a 96-well plate.

4 Add approximately 1mm^2 piece of tissue (usually operculum punch or adipose fin clip) in each well. Be careful when doing this, as it is easy to mess up.

5 Add **5 µL** of proteinase K to each well using a repeater pipette. This should always be the last step, as proteinase K will digest itself at room temperature with no substrate.

6 Cover plate and incubate in the thermocycler under the following conditions: 60°C for 120 min (tissue digestion) 95°C for 15 min (heat kill for proteinase K) This protocol should be saved in most of the thermocyclers (just scroll through). Sometimes this is not sufficient for complete digestion, so a second addition of 5uL of proteinase K followed by a repeat of the incubation procedure may be necessary. Alternatively, you could increase the digestion period from 120 min to 240 min. This is what I usually do and seems to work quite well.

7 Once digestion is complete, vortex briefly to break up any remaining pieces of tissue, then vortex at 1500 rpm for 2 min to collect Chelex and any other remaining solids on the bottom of the well.

8 Pipette off the supernatant and add to a new 96 well. This will be your undiluted DNA stock that will be stored for future use. Store at -20°C in chimney well plates with caps.

9 Make up a 1:10 dilution plate to be used for the assays. Usually **200 µL** is sufficient to run all assays. Add **180 µL** of dH2O to each well, and then add **20 µL** of your undiluted stock. Vortex and spin to finish the procedure. The 1:10 dilution plate can be stored in the fridge to avoid multiple freeze/thaw cycles.

Citation: Sarah Chang, Michael Russello Chelex DNA extraction Protocol https://dx.doi.org/10.17504/protocols.io.byvhpw36