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DNA extraction protocol for the Bungarus multicinctus by using AxyPrepTM Multisource Genomic DNA Miniprep Kit V.1

In 1 collection

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ABSTRACT

Genomic DNA was extracted from the muscle of a *Bungarus multicinctus* using the AxyPrep^{TM} Multisource Genomic DNA Miniprep Kit (Axygen, China) with modifications as outlined in this protocol.

Keywords: DNA extraction,
AxyPrepTM Multisource
Genomic DNA Miniprep Kit

1 The following protocol is a modification of the protocol by using the AxyPrep^{TM} Multisource Genomic DNA Miniprep Kit.

Approximately 10mg tissue was minced with sterilized scissors on ice.

On ice

2 Add 350 µl Buffer PBS and 15 µl SDS Lysis Buffer and shake for 30s. 30s

00:00:30

3 Add 150 µl Buffer C-L and 20 µl Proteinase K. Immediately vortex for 1 min to mix well. After brief centrifugation, incubate at 56°C for 10 min. 11m

00:01:00

56 °C 00:10:00

4 Add 350 µl Buffer P-D, vortex for 30s to mix well, and centrifuge at 12,000×g for 10 min. 30s

00:00:30

12000 rpm, 00:10:00

5 Place the DNA preparation tube in a 2 ml centrifuge tube, transfer the mixture in step 4 to the preparation tube, and centrifuge at 12,000×g for 1 min.

12000 rpm, 00:01:00

6 Discard the filtrate, put the preparation tube back into the original 2 ml centrifuge tube, add 500µl Buffer W1, and centrifuge at 12,000×g for 1 min.

12000 rpm, 00:01:00

7 Discard the filtrate, put the preparation tube back into the original 2 ml centrifuge tube, add 700µl Buffer W2, centrifuge at 12,000×g for 1 min.

12000 rpm, 00:01:00

7.1 In the same way, cleanse again with 700µl Buffer W2.

Note

1. Confirm that absolute ethanol has been added to Buffer W2 concentrate according to the volume specified on the reagent bottle.
2. Cleansing again with Buffer W2 can ensure that the salt is completely removed and eliminates the impact on the enzyme digestion reaction.

8 Discard the filtrate, put the preparation tube back into the original 2ml centrifuge tube, and centrifuge at 12,000×g for 1 min.

12000 rpm, 00:01:00

9 Place the DNA preparation tube in another clean 1.5 ml centrifuge tube, add 100-200μl Eluent or deionized water to the center of the preparation tube membrane, stand at room temperature for 1 min, and centrifuge at 12,000×g for 1 min to elute DNA. 1m

00:01:00 Room temperature

12000 rpm, 00:01:00