

RNA extraction from torafugu pufferfish with Minilys

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CONTEXT

Our laboratory focuses on the mechanisms involved in the biological processes in relation to energy metabolism, stress responses and cell signaling pathway for sustainable and effective utilization of aquatic resources. The extraction of RNA from livers or muscles of various fishes is a part of laboratory's daily work.

MATERIAL

- Minilys homogenizer.
- Precellys kit: 03961-1-002 (2.8 mm ceramic beads).
- Sample: 100 mg of liver or muscle cut into 7 mm cubic pieces of torafugu pufferfish (*Takifugu rubripes* 33cm, 750g) stored in RNAlater at -20°C.
- Buffer: 1 mL Lysis reagent of Qiagen RNeasy lipid tissue mini kit.

PROTOCOL

- Minilys: 5000 or 4000 rpm, 2x30sec (15sec break at RT).
- Precellys: 5000 rpm, 2x30sec (15sec break at RT).
- Analysis: denatured agarose gel (0.9%) electrophoresis stained with Sybr Green. The amount of RNA was determined using a spectrophotometer.

RESULTS

The quality of extracted RNA from torafugu pufferfish was good (Fig.1). For the torafugu pufferfish muscle which is exceptionally hard among the fish muscle, 5000 rpm 2x 30sec gave very good RNA yield (Table1). The 4000 rpm treatment was not enough (the homogenate looked somewhat granular).

The RNA concentration extracted from the fish liver using Minilys was somewhat smaller than the amount of RNA extracted using Precellys24.

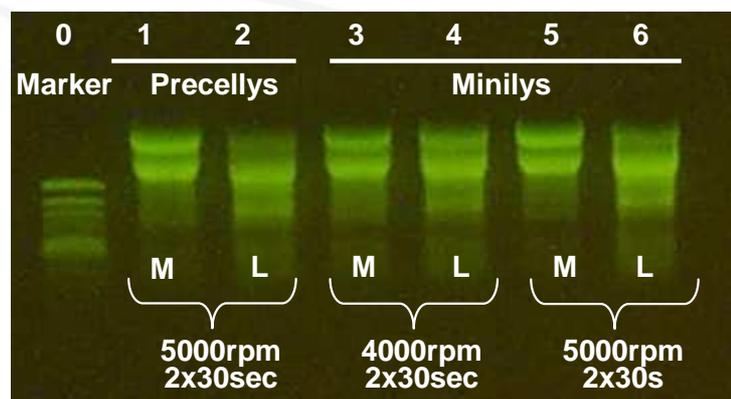


Figure 1: Agarose gel electrophoresis of the RNA extracted from torafugu pufferfish

	Precellys		Minilys	
	5000 rpm 2x30s	4000 rpm 2x30s	5000 rpm 2x30s	5000 rpm 2x30s
Pufferfish Muscle	1,40	2,04	2,73	
Pufferfish Liver	1,49	0,49	0,50	

Table 1: concentration of RNA extracted (µg/µl)

CONCLUSION

Precellys24 and Minilys successfully extracted RNA from torafugu pufferfish liver and muscle.

Minilys is a suitable homogenizer to extract RNA from fish tissues for research in marine biology. Minilys is a real alternative for standardization of the sample preparation for laboratories with a low or medium throughput of sample.

