



PROTEINS EXTRACTION FROM EYE TISSUES WITH 96 WELL-PLATE KITS AND PRECELLYS® EVOLUTION

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/ CONTEXT

In research on ophthalmology, mice are commonly used as models for retinal degeneration. Proteins expression analysis is needed to evaluate therapeutic agent efficiency. Concentrated protein extracts are difficult to obtain because of the small size of tissues. The ratio between volume for extraction and size of tissue is crucial for a sufficient yield. Currently, we are extracting proteins with a motorized pestle mixer, taking time and variability in extraction yield. The goal was to prepare a large number of sample with high reproducibility using a faster and easier methods.

/ MATERIALS

- Precellys® Evolution
- Precellys® lysing kit: CKMIX_WP KT03961-1-709.WP
- Sample: Neural Retina and eye cup (composed of Retinal pigmented Epithelium/Choroid/Sclera) isolated from eye mouse and directly frozen in nitrogen liquid
- Buffer: 50µl M-PER buffer (Thermo-Scientific)

/ PROTOCOL

Precellys® Evolution parameters:

For retina: 5500 rpm, 3x30s (30s break on ice)

For eye cup: 8 200 rpm, 3x30s (30s break on ice)

Motorized rotor 1,5ml pestle:

For retina: 1 min with up and down movements

For eye cup: After scissors disruption, 4 min with up and down movements

After homogenization, the tubes were centrifuged for 5min at 13000 rpm at 4°C. The supernatant was used to evaluate the quantity of extracted protein.

/ CONCLUSION

The Precellys® Evolution, combined with the dedicated Lysing Kit, the CKMix in 96 wells plate format efficiently grind and homogenizes different eye tissues such as retina and eye cup. In less than 2 minutes, this process enable a highly reproducible protein extraction yield, for a large number of samples with small amount and buffer and with no pre-grinding step.

The Precellys® offers the best solution to grind sample easily with the 96 well-plate format.

/ RESULTS

With Precellys® Evolution, the retina and eye cup lysates were obtained faster and easily than with pestle. The 96 well plate format is suitable for large number of sample in small lysis buffer volume. The level of proteins extracted was higher.

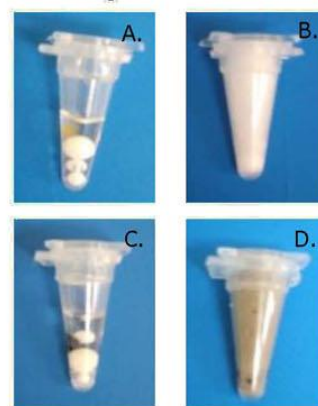


Fig 1: Homogenization of Retina and Eye cup using the CKMix 96 Well plate lysing kit

A. & B. Retina before and after homogenization

C. & D. Eye cup before and after homogenization

/ CUSTOMER

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【Precellys Evolution 対応 96 ウェルプレートキットを用いたマウス眼組織からのタンパク質抽出】 Precellys-APS-1704-89