

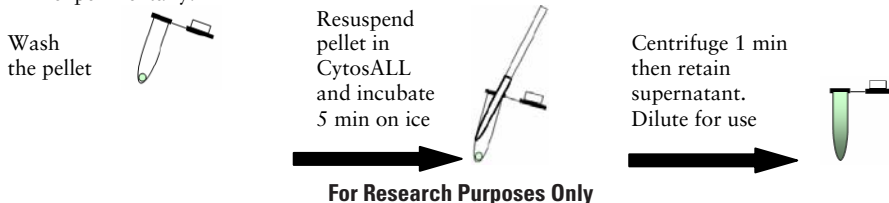
CytosALL™ Reagent

PRODUCT NUMBER: AB-4400
STORE: +4°C
SHIP: Ambient within the UK and wet ice for international and within the US.

INSTRUCTIONS FOR USE

Reagent for the preparation of cytoplasmic lysates for direct use in applications such as RT-PCR and QRT-PCR. RNA purification is not required. This protocol is recommended for adherent cells (grown in a monolayer) or cells grown in suspension.

- 1) Prepare cell pellet:
 - a. Harvest and pellet 1×10^6 cultured cells. Place on ice. Wash once with cold phosphate buffered saline (PBS).
 - b. Remove and discard supernatant PBS.
- 2) Prepare lysis reagent:
 - a. For each 1×10^6 cell pellet, add 6 μ l RNase inhibitor (20-40 units/ μ l) to 600 μ l CytosALL Reagent.
- 3) Lyse the cells:
 - a. Add 600 μ l cold lysis reagent (from step 2.a.) to the pellet.
 - b. Resuspend the pellet by pipetting eight to ten times or until the pellet is completely disrupted.
 - c. Incubate on ice for five minutes. Incubation times greater than 15 minutes may affect the yield of RNA.
- 4) Recover the cytoplasmic lysate:
 - a. Centrifuge the cell lysate at +4°C for one minute at 15,000 x g. While the cell lysate is centrifuging, place a new microcentrifuge tube on ice.
 - b. Immediately transfer the supernatant cytoplasmic lysate to the new chilled microcentrifuge tube.
- 5) Aliquot and store the undiluted lysate at -20°C or -80°C (for up to two weeks) or use directly in desired application.
 - a. RNA purification is not required for downstream applications, such as RT-PCR and QRT-PCR.
- 6) Some applications may require dilution of the lysate. Table 1 (overleaf) provides general recommendations for dilution; the optimal dilution will need to be determined experimentally.



	Suggested Dilution Range	Volume to add to 20 µl RT reaction	Volume to add to 25 µl PCR reaction	Cell equivalents
1-step RT-PCR	1:2 – 1:1,000	-	1 µl diluted sample	1 – 1,000
1-step QRT-PCR	1:75 – 1:15,000	-	5 µl diluted sample	<0.5 –100*
2-step RT-PCR	Neat – 1:20,000	1 µl diluted sample	1 – 2.5 µl RT reaction	1 –400
2-step QRT-PCR	1:5 – 1:50,000	1 µl diluted sample	1 –5 µl RT reaction	<0.5 – 50*

*For linear amplification (when absolute quantification is desired) use 0.5-10 cells for 1-step QRT-PCR or 1-10 cells for 2-step QRT-PCR.

Warranty

This product (“Product”) is warranted to operate or perform substantially in conformance with published Product specifications in effect at the time of sale, as set forth in the Product documentation, specifications and/or accompanying package inserts (“Documentation”) and to be free from defects in material and workmanship. Unless otherwise expressly authorized in writing, Products are supplied for research use only. No claim of suitability for use in applications regulated by FDA is made. The warranty provided herein is valid only when used by properly trained individuals. Unless otherwise stated in the Documentation, this warranty is limited to one year from date of shipment when the Product is subjected to normal, proper and intended usage. This warranty does not extend to anyone other than the original purchaser of the Product (“Buyer”).

No other warranties, express or implied, are granted, including without limitation, implied warranties of merchantability, fitness for any particular purpose, or non infringement. Buyer’s exclusive remedy for non-conforming Products during the warranty period is limited to replacement of or refund for the non-conforming Product(s).

There is no obligation to replace Products as the result of (i) accident, disaster or event of force majeure, (ii) misuse, fault or negligence of or by Buyer, (iii) use of the Products in a manner for which they were not designed, or (iv) improper storage and handling of the Products.

Ordering Information

AB-4400/A	CytosALL™ Reagent	60 ml (100 rxns)
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For Research Purposes Only

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Related Products

Plates:	ABgene® SuperPlate™ ABgene® Thermo-Fast
cDNA Synthesis Kit:	Verso™ cDNA Synthesis Kit
QPCR Kits:	ABsolute™ Blue QPCR Mix ABsolute™ Blue QPCR SYBR Green Mix
QRT-PCR Kits:	Verso™ 1-Step QRT-PCR Kit Verso™ SYBR® Green 1-Step QRT-PCR kit Verso™ 2-Step Kit Verso™ SYBR® Green 2-Step Kit

RNase Inhibitor: Porcine Ribonuclease Inhibitor

These products are available under multiple references and format to fit most of the instruments of the market. Please refer to our website www.abgene.com or contact us directly for more information (details below).

Troubleshooting

For troubleshooting, see www.abgene.com/troubleshoot.asp or contact Thermo Fisher Scientific (ABgene) TechSupport at abgene.techsupport@thermofisher.com

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