

(intype IC-DNA Cat. no. IC289980 or intype IC-RNA Cat. no. IC289970)

Products	Volume	Storage Conditions
intype IC-DNA: exogenous control DNA (2 x 10 ⁵ copies/μL)	1x1 mL	-30°C to -15°C
intype IC-RNA: exogenous control RNA (1 x 10 ⁵ copies/μL)	1x1 mL	-30°C to -15°C

Description

intype IC-DNA and intype IC-RNA are exogenous Internal Controls (IC), which can be used as extraction and amplification controls to exclude false-negative results caused by PCR inhibitors remaining after nucleic acid extraction. In addition, intype IC-DNA and IC-RNA monitor the entire molecular workflow for inhibition or other workflow issues, including mishandling, extraction, reagent or instrument errors and failures. The intype-IC products are designed to be sensitive to inhibition in the absence or presence of the main target sequence in the PCR. Significant Ct value shift of intype IC in negative samples indicates strong inhibition, which may yield false negative results.

- The intype IC-DNA or IC-RNA can be added to the lysis buffer for purification of nucleic acid from samples to serve as an extraction and amplification control, see **Protocol A**
- The intype IC-DNA or IC-RNA can be added to the PCR master mix to serve as only an amplification PCR control, see **Protocol B**

Storage and Handling

- Upon receipt, store the material at -30°C to -15°C, secured from any sources of contaminating DNA or RNA, especially amplified DNA
- Avoid repeated thawing and freezing (>5x) as this may reduce performance
- If used intermittently, aliquot the material and freeze
- Use aerosol barrier pipet tips for pipetting
- Do not use after the expiration date printed on the label

Pairing with INDICAL's IndiMixes (recommended)

intype IC-DNA and IC-RNA can be paired with IndiMix™ JOE (Cat. nos. MX299945, MX299947) or IndiMix TAMRA (Cat. nos. MX299985, MX299987) master mixes to enable monitoring of real-time amplification of single or multiple nucleic acid targets. These IndiMixes contain enzymes to amplify both DNA and RNA targets, and the primers and probe for intype IC-DNA and IC-RNA amplification. The IndiMixes are designed to work with fast cycling parameters and are compatible with standard real-time PCR instruments. IndiMixes can be purchased separately from INDICAL (contact details below).

Alternative master mix - ordering intype IC primers and probe

If using an alternative master mix, the intype IC primers and probe can be ordered separately and pipetted into the PCR mix following the manufacturer's instructions to enable detection of intype IC-DNA or IC-RNA. The concentration of the intype IC primers and probe depends on the PCR chemistry

used and the intended application in the laboratory. Primer and probe sequences are available on request from INDICAL Technical Support (contact details below).

Procedures

Protocol A: intype IC as an extraction and amplification control

The volume of intype IC products used during an extraction should be optimised to the extraction chemistry. intype IC-DNA or IC-RNA can be pipetted into the lysis buffer before adding to the samples. When the lysis procedure includes a heating step, the intype IC-DNA or IC-RNA should be added afterwards to prevent degradation of the controls. The input volume of intype IC depends on the final elution volume and the desired Ct value in the absence of inhibitors. We recommend starting with 1 µL of intype IC per sample in the lysis buffer for a 100 µL elution volume. If an earlier or later Ct value is desired, adjust the input amount of intype IC.

Add the intype IC to the extraction

1. Add 1 µL of intype IC per sample to the lysis buffer. Bulk volumes can be prepared if extracting multiple samples. If the intype IC Ct value is undesirable for the laboratory's application, the volume of intype IC added can be adjusted.

Note: If the lysis step involves heat, add the intype IC control after the heating step.

2. Proceed with the extraction according to the manufacturer's recommendations.

Nucleic Acid Amplification

1. Configure the real-time PCR instrument in accordance with the PCR chemistry manufacturer's thermocycling conditions.
2. Create the master mix according to the manufacturer's specifications, ensuring the primers and probe to detect the intype IC are included in the master mix (if not using IndiMix JOE/TAMRA).
3. Aliquot the master mix to the appropriate PCR wells for samples and extraction controls (if used), positive intype IC PCR control, positive target control, and negative PCR control containing nuclease-free water.
4. Prepare a positive intype IC PCR control by creating a 1:100 dilution of the intype IC control in RNase free water (1 µL of intype and 99 µL of RNase free water). Use a volume of this dilution equal to that of the eluted samples.
5. Add an equal volume of samples and controls to the designated PCR wells.
6. Load the PCR plate or tubes into the thermal cycler and run the manufacturer's protocol.

Protocol B: intype IC as a PCR amplification control

For samples extracted without IC, intype IC can be added directly in the PCR master mix and is compatible with IndiMix JOE, IndiMix TAMRA or with other PCR chemistries. The input volume of the intype IC-DNA or IC-RNA depends on the PCR chemistry and the desired Ct value in the absence of inhibitors. We recommend starting with 0.1 µL of undiluted intype IC per reaction or use a larger volume of a further diluted intype IC to achieve this concentration.

Nucleic acid amplification

1. Configure the real-time PCR instrument in accordance with the PCR chemistry manufacturer's thermocycling conditions.
2. Create the master mix according to the manufacturer's specifications and add 0.1 µL of the intype IC-DNA or IC-RNA per PCR reaction. Alternatively a 1:10 dilution of intype IC can be made (1µL of intype IC and 9µL of RNase free water) and add 1µL per PCR reaction, adjusting the final reaction volume as needed. If an earlier or later Ct value is desired, adjust the input amount of intype IC as required. Ensure that the primers and probe for detecting the intype IC are included in the master mix (if not using IndiMix JOE/TAMRA).
3. Aliquot the master mix to the appropriate PCR wells for testing nucleic acid samples, positive target control, and a negative PCR control containing nuclease-free water.

***Note:** The negative PCR control is not a true negative, as intype IC amplification is expected. Accordingly, this control serves both as an internal control and a no-target control in this system.*

4. Add an equal volume of samples and controls to the designated PCR wells.
5. Load the PCR plate or tubes into the thermal cycler and run the manufacturer's protocol.

Analyzing the results

Carefully examine the amplification plot, adjusting the baseline and threshold values as needed. Set the manual threshold setting at 5% of the maximum amplification from the positive intype IC PCR control (**Protocol A**) or negative PCR control (**Protocol B**).

The target and/or intype IC should amplify within each PCR reaction if no/minimal inhibitors are present. The intype IC control is designed to be secondary to the other targets and may not amplify in the presence of strong positive samples. The positive target result is valid in the absence of an intype IC signal. Suggested interpretation guidelines are provided below. Each laboratory should establish an interpretation of the intype IC results based on their applications.

Scenario	Target Result	intype IC result	Target Interpretation
One	Any Ct value	Any Ct value	Valid
Two	Any Ct value	No Ct value	Valid
Three	No Ct value	Any Ct value *	Valid
Four	No Ct value	No Ct value *	Invalid

*In scenario three, an intype IC Ct value >35 may indicate inhibition. Inhibition may also be present in scenario 4. In these incidences repeat the extraction, including a 1:1 dilution of the sample if possible, or repeat PCR amplification using a 5× dilution of the extracted nucleic acid in nuclease-free water.

For more information, please contact INDICAL Technical Support (contact details below).

Quality Control

Each lot of this product was manufactured and released in accordance with INDICAL's ISO-certified Quality Management System.

Safety Information

Please consult the appropriate safety data sheets (SDSs) for this product. For more information, the SDSs are available from your regional sales manager or can be provided by emailing compliance@indical.com. When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles.

Change index

Handbook	Version	Change
HB-2708-EN-001	March 2026	Launch new manual

For molecular biology applications. This product is not intended for the diagnosis, prevention, or treatment of a disease.

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