

LightCycler[®] Capillaries (20 μl)

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Reaction vessels for the LightCycler® 2.0 Instrument

Cat. No. 04 929 292 001 5 × 96 capillaries

containing 5 boxes, each with 96 capillaries and stoppers

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Store product at +15 to +25°C.

1.	General Information	3
1.1.	Contents	3
1.2.	Storage and StabilityStorage Conditions (Product)	
1.3.	Additional Equipment and Reagent required	3
1.4.	Application	3
2.	How to Use this Product	4
2.1.	Protocols	4
3.	Additional Information on this Product	
3.1.	Test Principle	6
3.2.	Quality Control	6
4.	Supplementary Information	7
4.1.	Conventions	
4.2.	Changes to previous version	7
4.3.	Trademarks	7
4.4.	License Disclaimer	7
4.5.	Regulatory Disclaimer	7
4.6.	Safety Data Sheet	8
4.7.	Contact and Support	8

1. General Information

1.1. Contents

Vial / Bottle	Label	Function / Description	Content
1	LightCycler [®] Capillaries (20 µl)	One box contains 96 capillaries	5 boxes

1.2. Storage and Stability

Storage Conditions (Product)

When stored at +15 to +25°C, the LightCycler® Capillaries (20 µl) are stable through the expiry date printed on the label.

1.3. Additional Equipment and Reagent required

For PCR reactions

- LightCycler® 2.0 Instrument
- the LightCycler® Centrifuge Adapters, in combination with a standard benchtop microcentrifuge, containing a rotor for 2.0 ml reaction tubes
- 1 The LightCycler® Carousel-Based System is provided with centrifuge adapters that allow LightCycler® Capillaries (20 μl) to be centrifuged in a standard microcentrifuge rotor.

or

- LC Carousel Centrifuge 2.0 for use with the LightCycler[®] 2.0 Sample Carousel (20 μl)
- To adapt the LightCycler® 2.0 Carousel (20 μl) to the former LC Carousel Centrifuge, you need the LC Carousel Centrifuge 2.0 Rotor Set.
- LightCycler® Capping Tool (included with the LightCycler® Carousel-Based System)
- LightCycler[®] 2.0 Capillary Releaser (included with the LightCycler[®] Carousel-Based System)

1.4. Application

The LightCycler® Capillaries (20 µl) are the appropriate reaction vessels for the LightCycler® Carousel-Based System. The capillary can hold reaction volumes ranging from 10 to 20 µl. High-quality borosilicate glass ensures superb PCR performance and optimal fluorescence transmission.

- The LightCycler® Capillaries (20 μl) are compatible with the LightCycler® 2.0 Sample Carousel (20 μl) and the LightCycler® Sample Carousel.
- If the LightCycler® Capillaries (20 μl) are used in combination with the LightCycler® 2.0 Instrument and Roche Diagnostics LightCycler® kits for in vitro diagnostic use (not available in all countries), adhere to the Instructions for Use of these kits.

2. How to Use this Product

2.1. Protocols

During set-up of a LightCycler® Carousel-Based System PCR run, PCR components (reagent mix and template nucleic acid, either separately or combined) are pipetted into the capillary reservoir and then forced into the glass capillary by centrifugation. You can perform this centrifugation step in two different ways:

- by using the LightCycler® Centrifuge Adapters, in combination with a standard benchtop microcentrifuge, or
- by using the LightCycler[®] 2.0 Sample Carousel (20 μl), in combination with the LC Carousel Centrifuge 2.0.
- Do not touch the surface of the capillaries. Always wear gloves when handling the capillaries and the stoppers.
- 1 Program the experimental protocol and define the sample numbers, names etc. Refer to the 'Software' section of the LightCycler® 2.0 Instrument Operator's Manual for detailed instructions.
 - Mhen programming the experimental protocol, ensure that you select "20 μl" as the Capillary Size for the experiment.
- 2 Prepare a PCR reagent mix and omit the template nucleic acid. Details on preparing the PCR mix are described in the corresponding Instructions for Use of the LightCycler® reagent kit in use.
- 3 Mix by gentle vortexing.
- Place the capillaries into the LightCycler® Centrifuge Adapters that have been precooled in the cooling block.
 Pipette the PCR mix into the plastic reservoir at the top of the capillary. Add the template nucleic acid to the capillary.
- 5 Seal each capillary with a plastic stopper using the LightCycler® Capping Tool.
 - A Ensure that each LightCycler® Capillary (20 μl) is closed tightly by checking it visually: The lower part of the plastic stopper must be completely inserted into the glass capillary.
- 6 Perform one of two different centrifugation steps:

Preparing a PCR run using the LightCycler® Centrifuge Adapters

- Place the centrifuge adapters each containing a LightCycler[®] Capillary (20 μl) into the rotor of a standard benchtop microcentrifuge.
- Use only rotors that are designed to hold 2.0 ml reaction tubes.
- A Place the centrifuge adapters in a balanced arrangement within the centrifuge. If centrifuging an uneven number of capillaries, use an empty centrifuge adapter as counterweight.
- Centrifuge briefly, at not more than 735 \times g for 5 s.
- Place the capillaries into the LightCycler® 2.0 Sample Carousel, keeping the capillaries in an upright position.
- Make sure that all capillaries are fixed in the optimal position where the O-ring of the LightCycler® 2.0 Sample Carousel covers the lower part of the plastic reservoir. Ensure proper positioning by lightly pressing the cap until you hear a final "click" as the capillary reaches its final position.
- ⚠ When pressing the capillaries into the LightCycler® 2.0 Sample Carousel, do not press too hard. Do not use capillaries that show slight cracks or cracked slightly when inserted into the LightCycler® 2.0 Sample Carousel. Do not use capillaries that have been dropped. Capillaries with slight cracks, possibly invisible, might rupture during a run.

Preparing a PCR run using the LC Carousel Centrifuge 2.0

- Place the capillaries into the LightCycler® 2.0 Sample Carousel, keeping the capillaries in an upright position.
- ⚠ Make sure that all capillaries are fixed in the optimal position where the O-ring of the LightCycler®
 2.0 Sample Carousel covers the lower part of the plastic reservoir. Ensure proper positioning by
 lightly pressing the cap until you hear a final "click" as the capillary reaches its final position.
- Mhen pressing the capillaries into the LightCycler® 2.0 Sample Carousel, do not press too hard. Do not use capillaries that show slight cracks or cracked slightly when inserted into the LightCycler® 2.0 Sample Carousel. Do not use capillaries that have been dropped. Capillaries with slight cracks, possibly invisible, might rupture during a run.
- Put the loaded LightCycler® 2.0 Sample Carousel into the rotor bucket and place it in the LC Carousel Centrifuge 2.0.
- Only operate the LC Carousel Centrifuge 2.0 with the blue rotor and blue rotor bucket supplied together with the instrument.
- <u>Λ</u> To adapt the LightCycler® 2.0 Sample Carousel (20 μl) to the former LC Carousel Centrifuge, you need the LC Carousel Centrifuge 2.0 Rotor Set.
- 7 Refer to the LC Carousel Centrifuge 2.0 Operator's Manual for detailed operating instructions.
- Place the sample carousel into the LightCycler® Carousel-Based System Instrument. Ensure that the notch below sample position 1 on the sample carousel locks into position against the pin on the thermal chamber. Visually check that the carousel is inserted correctly and fits perfectly in the thermal chamber.
 - ⚠ Before placing the sample carousel into the LightCycler® Carousel-Based System:
 - Visually check that all capillaries have a uniform fill level or if capillary breakage has occurred. Any indications of an uneven fill level will point to possible leakage of a capillary. Discard the affected capillaries, as leakage could impair the experimental result especially in quantification experiments.
 - If capillary leakage or breakage occurs, clean the LightCycler® Centrifuge Adapters or the sample carousel with a lab tissue soaked with decontamination solution. Use commercialized reagents such as LTK-008 (Biodelta) or DNAZap (Ambion). The sample carousel can also be autoclaved. Clean the chamber of the benchtop centrifuge or the LC Carousel Centrifuge 2.0 with a mild commercial detergent. If necessary, use 70% ethanol for disinfecting the chamber. Refer to the LightCycler® 2.0 Instrument Operator's Manual and the LC Carousel Centrifuge 2.0 Operator's Manual for detailed instructions.
 - Ensure that the thermal chamber is clean and free of any items that could interfere with the capillaries during the run.
- 8 Close the instrument lid. You are now ready to start the run.
- 9 At the end of the run, release all capillaries placed into the sample carousel using the LightCycler® 2.0 Capillary Releaser:
 - Place the loaded sample carousel on the capillary releaser, then push the carousel down with the flat of your hand. The capillaries will be released from the rubber O-ring and held at a slightly elevated position. You can now easily remove the capillaries from the sample carousel.
- 10 After use, discard the capillaries into a solid waste box.

3. Additional Information on this Product

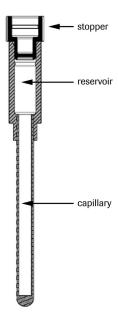
3.1. Test Principle

In the LightCycler® Carousel-Based System, temperature cycling is achieved using air. As air has virtually no mass, this process is significantly faster (ten times) than cycling with conventional thermal blocks. A heating coil controlled by thermocouples in the thermal chamber of the LightCycler® Carousel-Based System warms the air. PCR occurs in the specially designed borosilicate glass LightCycler® Capillaries. The capillaries have a high surface-to-volume ratio, to ensure rapid equilibration between the air and the reaction components.

The combination of using air for rapid thermal cycling and the high surface-to-volume ratio of the capillaries, enables a single PCR cycle to be completed in less than 50 seconds using the LightCycler® Capillaries (20 µl). Depending on the experimental protocol, a complete PCR run of 30 to 40 cycles can thus be performed in 25 to 35 minutes on the LightCycler® Carousel-Based System.

Background information

The optical properties of borosilicate glass make the capillaries ideally suited for use as cuvettes for fluorescence measurement. A glass capillary is molded to a polypropylene reservoir, permitting easy loading of the capillary. A stopper provides a secure seal, significantly reducing the risk of contamination. Each LightCycler® Capillary (20 µl) is 45 mm long (without plastic stopper) and has an outer diameter of 1.55 mm.



3.2. Quality Control

Each lot of LightCycler® Capillaries (20 µl) is function tested using the LightCycler® Carousel-Based System.

4. Supplementary Information

4.1. Conventions

To make information consistent and easier to read, the following text conventions and symbols are used in this document to highlight important information:

Text convention and symbols					
1 Information Note: Additional information about the current topic or procedure.					
⚠ Important Note: Information critical to the success of the current procedure or use of the product.					
1 2 3 etc.	Stages in a process that usually occur in the order listed.				
1 2 3 etc.	Steps in a procedure that must be performed in the order listed.				
* (Asterisk)	The Asterisk denotes a product available from Roche Diagnostics.				

4.2. Changes to previous version

Layout changes.

Editorial changes.

Change of the regulatory disclaimer to "For general laboratory use".

4.3. Trademarks

LIGHTCYCLER is a trademark of Roche.

All other product names and trademarks are the property of their respective owners.

4.4. License Disclaimer

For additional documentation such as certificates and safety data sheets, please visit: **documentation.roche.com**.

4.5. Regulatory Disclaimer

For general laboratory use.

4.6. Safety Data Sheet

Please follow the instructions in the Safety Data Sheet (SDS).

4.7. Contact and Support

If you have questions or experience problems with this or any Roche product for Life Science, please contact our Technical Support staff. Our scientists are committed to providing rapid and effective help.

Please also contact us if you have suggestions for enhancing Roche product performance or using our products in new or specialized ways. Such customer information has repeatedly proven invaluable to the research community worldwide.

To ask questions, solve problems, suggest enhancements or report new applications, please visit our **Online Technical Support** Site.

Visit <u>documentation.roche.com</u>, to download or request copies of the following Materials:

- Instructions for Use
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