

QX200[™] Droplet Generation Oil for EvaGreen

Catalog # Description

1864005 **QX200 Droplet Generation Oil for EvaGreen**, 14 ml (2 x 7 ml vials), enough for 192 x 20 μl reactions **QX200 Droplet Generation Oil for EvaGreen**, 70 ml (10 x 7 ml vials), enough for 960 x 20 μl reactions

For research purposes only.

Description

QX200 Droplet Generation Oil for EvaGreen is used to create water (sample) in oil droplets required for Droplet Digital™ PCR (ddPCR™) analysis and is formulated for use exclusively with QX200 ddPCR EvaGreen Supermix (catalog #1864033, 1864034, 1864035, and 1864036).

Following reaction preparation using QX200 ddPCR EvaGreen Supermix, 20 µl each of up to eight prepared samples (or blanks) and QX200 Droplet Generation Oil for EvaGreen are transferred to a DG8™ Droplet Generator Cartridge. The loaded cartridge is covered with a DG8 Gasket and placed in the QX200 Droplet Generator. There, the samples and oil are combined within the microchannels of the cartridge to create an emulsion of ~20,000 monodispersed, nanoliter-sized droplets for each of the samples.

Additional requirements for droplet generation are listed in Table 1. For complete system requirements, refer to the QX200™ Droplet Reader and QuantaSoft™ Software Instruction Manual (#10031906).

Storage and Stability

QX200 Droplet Generation Oil for EvaGreen is stable for 12 months when stored at a constant temperature of 25°C (room temperature).

Sample Preparation

Prepare the PCR reaction as described in the QX200 ddPCR EvaGreen Supermix Product Insert (#10028376).

Droplet Generation

The QX200 Droplet Generator prepares droplets for up to eight samples at a time. Droplet generation takes ~2 min for each set of eight samples (~45 min for a full 96-well plate). The detailed protocol for droplet generation can be found in the QX200 Droplet Generator Instruction Manual (#10031907).

Note: QX200 Droplet Generation Oil for EvaGreen should be used only with QX200 ddPCR EvaGreen Supermix. The use of other supermixes can lead to poor results and potential damage to the QX200 Droplet Reader.

Table 1. QX200 droplet generation components.

| Component | Description | Catalog # |
|--|---|---|
| QX200 Droplet Generator | Instrument used for droplet generation | 1864002 |
| Droplet generator cartridges and gaskets | Microfluidic DG8 Cartridge used to mix sample and oil to generate droplets; DG8 Gaskets seal the cartridge to prevent evaporation and apply the pressure required for droplet formation | 1864007 |
| DG8 Cartridge Holder | Positions and holds the DG8 Cartridge in the instrument for droplet generation | 1863051 |
| PCR supermix | QX200 ddPCR EvaGreen Supermix | 1864033, 1864034, 1864035, 1864036 |
| Control | QX200 Buffer Control for EvaGreen | 1864052 |
| Pipets | 20 µl pipet for sample loading 50 µl pipet for droplet transfer 8-channel, 200 µl pipet for oil | Rainin L-20 Rainin L50, L8-50 Rainin L8-200 |
| Pipet tips | Filtered | Rainin GP-L10F, GP-L200F |
| 96-well PCR plates | twin.tec 96-well semi-skirted plates | Eppendorf 951020362 |
| Reagent trough | Any | |
| Foil seal | Pierceable foil heat seal | 1814040 |
| Plate sealer | PX1 [™] PCR Plate Sealer | 1814000 |
| 8-cap strips | Any | |

- 1. Insert the DG8 Cartridge into the holder, aligning the notch in the cartridge with the upper left corner of the holder.
- 2. Transfer 20 µl of each prepared sample to the sample wells (middle row) of the DG8 Cartridge.
- 3. Dispense the QX200 Droplet Generation Oil for EvaGreen into a reagent trough (see Table 2 for volumes required).
- 4. Using a multichannel pipet, fill each oil well (bottom row) with 70 μ l of QX200 Droplet Generation Oil for EvaGreen from the reagent trough.
- Hook the gasket over the cartridge holder using the holes on both sides. The gasket must be securely hooked on both ends of the holder to ensure pressure is sufficient to generate droplets.
- 6. Open the QX200 Droplet Generator by pressing the button on the green top and place the cartridge holder into the instrument.
- 7. Press the button on the top again to close the door and initiate droplet generation.
- When droplet generation is complete, all three indicator lights are solid green and the door can be opened by pressing the button. The holder, with the DG8 Cartridge still in place, can be removed.
- Pipet 40 µl of the contents of the top wells (the droplets) into a single column of a 96-well PCR plate. Repeat for each cartridge, filling up each column across the 96-well plate.
- To avoid evaporation, seal the PCR plate with foil immediately after transferring all the columns of droplets.
- 11. Begin thermal cycling (PCR) within 30 min of sealing the plate, or store the plate at 4°C for up to 4 hr prior to thermal cycling. Refer to the QX200 ddPCR EvaGreen Supermix Product Insert for cycling conditions.

Table 2. QX200 Droplet Generation Oil requirements.

| <u>.</u> | | |
|-----------------|-------------------|--|
| Number of Wells | Volume of Oil, μl | |
| 8 | 700 | |
| 24 | 1,820 | |
| 48 | 3,500 | |
| 96 | 6,860 | |

When PCR amplification is complete, remove the 96-well plate from the thermal cycler and read the droplets using the QX200 Droplet Reader (follow the directions in the QX200 Droplet Reader and QuantaSoft Software Instruction Manual).

For more information, visit

www.bio-rad.com/web/DGOilEvaGreen.

Eppendorf and twin.tec are trademarks of Eppendorf AG. EvaGreen is a trademark of Biotium, Inc. Bio-Rad Laboratories, Inc. is licensed by Biotium, Inc. to sell reagents containing EvaGreen Dye for use in real-time PCR, for research purposes only. Rainin is a trademark of Rainin Instrument, LLC.

The QX100 or QX200 Droplet Digital PCR System and/or its use is covered by claims of U.S. patents, and/or pending U.S. and non-U.S. patent applications owned by or under license to Bio-Rad Laboratories, Inc. Purchase of the product includes a limited, non-transferable right under such intellectual property for use of the product for internal research purposes in the field of digital PCR only. No rights are granted for diagnostic uses. No rights are granted for use of the product for commercial applications of any kind, including but not limited to manufacturing, quality control, or commercial services, such as contract services or fee for services. Information concerning a license for such uses can be obtained from Bio-Rad Laboratories. It is the responsibility of the purchaser/end user to acquire any additional intellectual property rights that may be required.