February 2025

Quick-Start Protocol

QIAprep&amp® Plasmodium Kit DBS Direct Workflow

The QIAprep&amp Plasmodium Kit (cat. no. 223213) should be stored immediately upon receipt at –30 to –15°C in a constant-temperature freezer and protected from light. Several components of the kit can be stored at room temperature (15–25°C): Blood Lysis Buffer, PR Buffer, DBS Wash Buffer. The S-Solution is stored light-protected at –20°C. This kit can be used for the detection of the Plasmodium parasite (in combination with the respective assay) in different workflows. The components used differ between workflows.

This workflow is designated to be used with dried blood spots (DBS) directly in the reaction; it is intended to be used with whole blood samples collected as DBS.

Further information

- *QIAprep&amp Plasmodium Kit Handbook*: **www.qiagen.com/HB-3663**
- Safety Data Sheets: **www.qiagen.com/safety**
- Technical assistance: **support.qiagen.com**
- *Pf/Non-Pf Detection Assay Kit Quick-Start Protocol*: **www.qiagen.com/HB-3669**
- *Pv/Pm/Po/Pk Detection Assay Kit Quick-Start Protocol*: **www.qiagen.com/HB-3671**

- *QIAprep&amp Plasmodium Kit Punching of Dried Blood Spots Quick-Start Protocol*: **www.qiagen.com/HB-3666**

Notes before starting

- The following blood collection cards are compatible: Whatman® filter paper (GE, cat. no. 3030-917 or 3017-915); QIAcard® Bloodstain card (cat. no. WB100014) or any other blood collection cards (untreated, no preservatives); as well as the QIAcard FTA Classic card (cat. no. WB 120205 or WB120305).
- It is recommended to use a puncher such as the Uni-Core® Punch 1.2 mm (cat. no. WB100074), or other comparable punchers.
- It is recommeded to use a Cutting mat (cat. no. WB100088).
- Thaw the QP&A DNA Mastermix, the PCR assay of choice, and RNase-free water.
- Additionally, this protocol uses S-Solution.
- Use the cycling conditions specified in this protocol.

Procedure

1. Vortex and centrifuge all reagents.
2. Take 1 paper punch from the blood sample collection card (preferably the middle of the blood spot) using a punching device (1.2 mm diameter of punch). Release the punch into the very bottom of a PCR plate/strip tube.

   Note: Go to **www.qiagen.com/HB-3666** for a detailed instruction on how to use the puncher.

   Important: Depending on the applied pressure, pieces of the mat can be transferred into the reaction. This does not have a negative effect on the result, however, coring of the mat should be avoided.

3. **PCR setup**: Prepare the PCR reaction mix for a multiplex PCR reaction as shown in Table 1. Vortex briefly and centrifuge to remove the liquid from the cap.

Table 1. Reaction mix setup

Component	Channel for detection	1 rxn (µL)	Final Concentration
QP&A DNA Mastermix	–	9	1x
20x Assay Mix	Select respective channels	1	1x
S-Solution	–	2	–
RNase-free Water	–	8	–
Total volume	**–**	**20**	**–**

4. Add 20 µL of PCR reaction mix (Table 1) to each well.
5. Seal the plate/tube thoroughly with a fresh foil/lid. Mix gently by vortexing with medium pressure (5–10 s). Place the plate in different positions while vortexing, to ensure an equal contact with the vortex platform.
6. Centrifuge the plate/tube briefly to collect the liquid at the bottom of the plate/tube. Place it in the real-time cycler and start the cycling program (with heated lid). Program the cycler as referred to in Table 2.

   Note: Data acquisition should be performed during the annealing/extension step.

Table 2. Cycling conditions

Step	Time	Temperature (°C)	Ramp rate
PCR initial heat activation	2 min	95	Maximal/fast mode
2-step cycling (40 cycles)			
Denaturation	5 s	95	Maximal/fast mode
Combined annealing/extension*	30 s	58	Maximal/fast mode

*Add data acquisition

Result interpretation

Refer to the following when using these assay kits:

a. *Pf/Non-Pf Detection Assay Kit Quick-Start Protocol*: **www.qiagen.com/HB-3669**

b. *Pv/Pm/Po/Pk Detection Assay Kit Quick-Start Protocol*: **www.qiagen.com/HB-3671**

For general qPCR result interpretation, refer to the *QIAprep&amp Plasmodium Kit Handbook* at **www.qiagen.com/HB-3663**

Document Revision History

Date	Changes
02/2025	Initial release

Scan QR code for handbook.

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual.

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