

Quick-Start Protocol

QIAprep&® Plasmodium Kit Punching of Dried Blood Spots

This protocol describes how to generate punches from dried blood spots (DBS). For the generation of dried blood spots we recommend: the QIAcard® Bloodstain card (cat. no. WB100014); or the QIAcard FTA Classic card (cat. no. WB 120205 or WB120305); or Whatman® filter cards (GE, cat. no. 3030-917 or 3017-915).

As puncher, we recommend Uni-Core[®] Punch 1.2 mm for DBS direct WF (cat. no. WB100074); and 3 mm for DBS Elution WF (cat. no. WB100078), or comparable punchers. Additionally the cutting mat (cat. no. WB100088) is required.

Further information

- QIAprep& Plasmodium Kit Handbook: www.qiagen.com/HB-3663
- Safety Data Sheets: www.qiagen.com/safety
- Technical assistance: support.qiagen.com
- QIAprep& Plasmodium Kit DBS Direct Workflow Quick- Start Protocol: www.qiagen.com/HB-3667
- QIAprep& Plasmodium Kit DBS Elution Workflow Quick- Start Protocol: www.qiagen.com/HB-3668

- 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

Notes before starting

- Use only DBS that are dry. When using freshly generated DBS, it is recommend to let the blood on the card dry for at least 6 hours (optimally overnight) before punching.
- Release each punch from the puncher tip before cutting a new punch. Do not cut several punches without ejecting.

• Always decontaminate the device to avoid contamination in the following order:

First, punching sample; second, release the punch from the device; and third, decontaminate the device. Follow the decontamination guidelines below between punches

to avoid transfer of nucleic acids. Ensure to decontaminate the punching mat between samples.

• Important: Dried blood spots have an unequal distribution of nucleic acids within the blood spot. It is recommended to punch in the middle of the DBS where the DNA concentration is the highest. If several paper punches are needed from one DBS sample, a slight variation among the Ct values can be observed due to the unequal distribution of DNA in the DBS.

Punching Procedure

- 1. Place the cutting mat on a lab bench. Position the paper card with the DBS on it.
- $2. \ \ \text{Remove the protective cap from the punch tip through gentle twisting}.$
- 3. Hold the puncher firmly between the fingers. Firmly press down the paper card with the other hand.
- 4. Place the punch tip vertically on the area (90° angle) in the middle of the blood spot. Apply gentle pressure on the spot and turn the puncher until it has successfully cut through

the paper card.

Important: Do not press the button at the top side of the device. Do not use too much force as the puncher is a delicate tool and might get damaged.

- 5. After cutting, the paper punch is fastened in the punch tip.
- 6. Eject the paper punch by placing the puncher with the tip into the tube or well, and lower the device down to the very bottom of the tube/well. Once lowered, press the button at the back end of the puncher to eject the punch.

Note: To avoid that paper punches stick to the side of a tube or well, put the puncher tip as deep as possible into the plasticware in use before ejecting the punch.

Decontamination Procedure

After each cut the puncher needs to be decontaminated:

- 1. Set up three small glass/plastic containers for the punchers to stand in during decontamination.
- 2. Prepare sufficient amounts of 10% bleach, distilled water, and 70% Ethanol:
 - a. Add 10% bleach to the first container (the volume should cover the needle of the puncher).
 - b. Add distilled water to the second container.
 - c. Add 70% Ethanol to the third container.

Important: Ensure that the Ethanol container (third) has the highest volume, then the distilled water container (second) has a lower volume, then the bleach container (first) has the lowest volume. This ensures the liquid of preceding containers will be completely washed away.

- 3. Insert the used puncher upright into the bleach container (so that it would stand), and the punching needle is completely immersed in the bleach. Incubate for 30 s.
- 4. Move the puncher into the distilled water container. Incubate for 30 s.
- 5. Move the puncher into the Ethanol container. Incubate for 30 s.
- 6. Dry the puncher with a lint-free tissue. Ensure that there are no Ethanol residues before punching the next sample.

The mat can be decontaminated by wiping with 10% bleach and subsequently with distilled water.

Note: Use multiple punchers in one experiment to avoid waiting times due to the decontamination procedure.

Document Revision History

Date	Changes
02/2025	Initial release



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