sparQ Lysis Kit

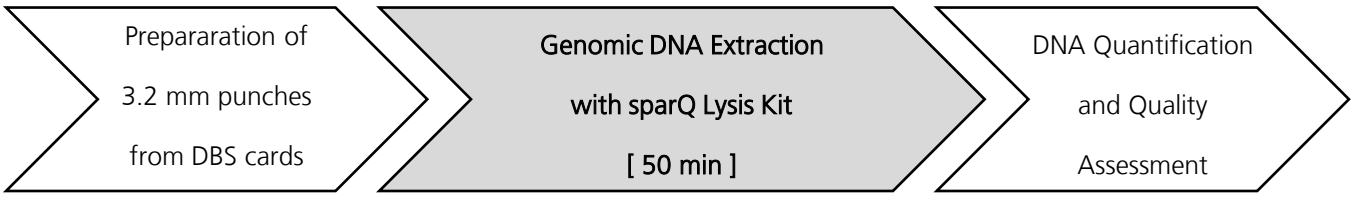
Cat. No.	95220-024 95220-096	Size	24 reactions 96 reactions	Store at room temperature (15°C to 25°C)

Description

The sparQ Lysis Kit includes reagents optimized for lysis of whole blood cells on dried blood spot (DBS) filters and the purification of high-quality genomic DNA (gDNA). The kit enables rapid extraction of high-molecular-weight gDNA (>50 kb) from DBS, whole blood, saliva, or cheek swab in just 50 minutes. The streamlined protocol supports both manual and automated, high-throughput workflows. Extracted DNA is suitable for PCR-free NGS, qPCR, dPCR, and targeted NGS applications.

The sparQ Lysis Kit is a key component of the sparQ DBS-seq Workflow, an end-to-end solution from DBS to sequencing-ready whole genome libraries. This Workflow is suitable for whole genome sequencing (WGS) application and supports both PCR-free or PCR-amplified library construction methods. As part of the Workflow, the sparQ Lysis Kit delivers intact, double-stranded gDNA for next-generation sequencing, even from limited sample input. The complete Workflow includes gDNA extraction, library preparation and library quantification, providing a streamlined and scalable solution.

Workflow Overview



Components

	Volume	
Component Description	24 reactions	96 reactions
DBSL Buffer	1 x 1.2 ml	4 x 1.2 ml
Proteinase K	1 x 240 µl	1 x 960 µl
Elution Buffer	1 x 1.32 ml	4 x 1.32 ml

Storage and Stability

Store the components at room temperature (15°C - 25°C) upon receipt.
For lot specific expiry date, refer to package label, Certificate of Analysis or Product Specification Form.

Instrument Requirement

The use of a thermomixer (Eppendorf ThermoMixer® C, Eppendorf ThermoMixer® F2.0, or equivalent) is essential for optimal extraction performance.

Additional reagents that are not supplied

- Nuclease-free water
- 80% freshly prepared ethanol
- Purification Beads for the cleanup step are not included with the kit and must be purchased separately. We recommend using sparQ PureMag Beads from Quantabio.

Part Number	Description	Kit Size
95196-005	sparQ PureMag Beads	5 ml
95196-060	sparQ PureMag Beads	60 ml
95196-450	sparQ PureMag Beads	450 ml

- To measure genomic DNA concentration, we recommend using Qubit® or equivalent method.
- To check genomic DNA quality, we recommend using Genomic DNA ScreenTape® in TapeStation® (Agilent) or equivalent method.

General Guidelines

- Use good laboratory practice to minimize cross-contamination of nucleic acid products.
- Always use PCR tubes, plates, microfuge tubes, and pipette tips that are certified sterile, DNase- and RNase-free.
- For consistent results, ensure the thermomixer used in this protocol is in good working order and has been calibrated to within the manufacturer's specifications.
- Briefly centrifuge tubes prior to opening to avoid loss of material.
- Read the entire protocol before beginning. Take note of stopping points and plan your workflow accordingly.

Point in protocol where procedure can be stopped, stored at appropriate conditions outlined, and continued within 24 hours

Take note of recommendations in protocol

Use caution when performing protocol to obtain the best results

Before You Begin

- Wipe down work areas and pipettes with an RNase and DNA cleaning product.
- Prepare 80% ethanol and bring the beads to room temperature at least 30 minutes before the experiment.
- Mix DBSL Buffer by inverting the tube 3 times. If any precipitation is visible, warm DBSL Buffer at 50°C for 15 minutes.

Protocol

DBS Processing: DNA Extraction from Dried Blood Spots

1. Program the thermomixer to 50°C and at 1000 rpm.
2. Bring sparQ PureMag Beads to room temperature 30 minutes before using.
3. Prepare 3.2-mm punches from DBS cards. Place punches in a 1.5 ml or 2 ml tube with rounded bottom.
4. Prepare Lysis Master Mix according to Table 1.

Note: The volumes indicated in Table 1 are per sample and can be scaled accordingly based on the number of samples being processed.

Table 1: Lysis Master Mix

	Per Sample Volume based on Number of Punches	
Component	1-5 Punches (µl)	6-10 Punches (µl)
DBSL Buffer	30	50
Proteinase K	5	10
Nuclease-free water	265	440
Total	**300**	**500**

5. Add the appropriate total 'per sample volume' of the Lysis Master Mix to the DBS punches.

Note: After adding reagents, vortex for 5 seconds and briefly centrifuge to collect contents at the bottom of the tube.

6. Incubate the DBS punches with the Lysis Master Mix on the preheated thermomixer at 50°C for 30 minutes. Shake at 1000 rpm.
7. Briefly centrifuge the samples to collect liquids.
8. Carefully transfer the supernatant (~290 or ~480 µl) to a fresh 2 ml tube.

Note: Ensure that the sparQ PureMag Beads has been kept at room temperature (RT) for at least 30 min before use. Thoroughly vortex the sparQ PureMag Bead slurry before adding.

9. To the lysed sample, add the sparQ PureMag Beads according to the below:
   a. For 1 - 5 punches: 120 µl
   b. For 6 - 10 punches: 200 µl
10. Mix the beads and sample thoroughly.
11. Incubate the mixture for 10 minutes at room temperature, preferably with shaking at 600 rpm.
12. Place the tubes on the magnetic stand, and incubate at room temperature for 5 minutes.
13. Pellet the beads on a magnetic stand and carefully remove and discard the supernatant.

14. Keeping the tube on the magnetic stand, gently pipette 500 µl of the freshly prepared 80% ethanol to wash the beads. Pellet the beads on the magnetic stand for 30 seconds then carefully remove and discard the supernatant. Repeat step 14 for a total of two washes.
15. Use a 10 µl pipette to remove any excess ethanol from the bottom of the tube.
16. Air-dry the beads on the magnetic stand for 3 minutes or until the beads appears matte and no longer shiny. Over-drying of beads may results in lower DNA recovery.
17. Remove the tube from the magnetic stand, resuspend the dried beads in **53 µl of Elution Buffer** and mix well by pipetting up and down at least 5 times. Incubate the beads at room temperature for 3 minutes. Pellet the beads on the magnetic stand. Carefully transfer **50 µl** of supernatant into a new thin-walled PCR tube.
18. Measure DNA concentration using Qubit or an equivalent method.
19. Genomic DNA quality can be checked using automated electrophoresis methods suitable for genomic DNA such as the Genomic DNA ScreenTape on the TapeStation instrument or equivalent.
20. Keep DNA on ice for immediate use or store at -20°C for long term storage.

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NGS Products Sold Separately for Use in the sparQ DBS-seq Workflow

sparQ DNA Frag & Library Prep Kit

Cat. No.	95194-024	Size:	24 reactions
	95194-096		96 reactions

The sparQ DNA Frag & Library Prep Kit is optimized for enzymatic fragmentation of DNA and streamlined construction of high-quality libraries for sequencing on NGS platforms. The simple, convenient 2-step workflow can be completed in 2.5 hours with minimal hands-on time and accommodates DNA input amounts from 1 ng to 1000 ng.

sparQ PureMag Beads

Cat. No.	95196-005	Size:	5 ml
	95196-060		60 ml
	95196-450		450 ml

The sparQ PureMag Beads is a fast and reliable nucleic acid purification system for reaction cleanup and size selection in NGS workflows. It can be used to quickly remove primers, primer-dimers, unincorporated nucleotides, salts, adapters and adapter-dimers from NGS library prep reactions to improve downstream sequencing performance.

sparQ UDI Adapters (1 – 96)

Cat. No.	95211-096	Size:	1 - 96

The sparQ UDI Adapters are unique dual-indexed barcoded adapters for DNA and RNA libraries for Illumina® sequencing platforms. It allows flexible pooling with improved performance by preventing index hopping and enhancing demultiplexing accuracy. The adapters are compatible with both DNA and RNA NGS libraries.

sparQ Universal Library Quant Kit

Cat. No.	95210-100	Size:	100 reactions
	95210-500		500 reactions
	95210-15B (dilution buffer)		15 ml

The sparQ Universal Library Quant Kit is optimized for rapid, sensitive, and accurate quantification of NGS libraries of various sizes and GC-contents. The kit uses fast cycling protocol, allowing results to be achieved in 40 minutes versus 1 hour and 20 minutes with other NGS library quantification kits.