

AutoGenFlex STAR Application Guide

1-2 ml Whole Blood



I. **Purpose:** To extract genomic DNA from 0.5 - 2.0 ml of EDTA, or citrate treated fresh or frozen whole blood.

II. **Chemical Principal:** QIAGEN's FlexiGene Procedure (protease digestion/DNA precipitation method)

III. Protocol Parameters:

A. **Sample Volume:** 0.5 - 2.0 ml of EDTA or citrate treated whole blood.

B. **Number of Samples:** Up to 40 samples per run.

C. **Throughput:** Up to 80 x 1-2 ml samples per day.

D. **Yield:** 12-36 µg of purified DNA per 1.0 ml of whole blood

E. **Quality:** Typical OD260/280 values are 1.70 – 1.90. The DNA is free of contaminants and inhibitors (RNA, protein) and can be used directly in downstream processes such as DNA sequencing, PCR-based assays, restriction digestions, and Southern blotting.

IV. Running the Protocol:

A. Load Reagents and Samples

B. Select the 1-2 ml Fresh/Frozen Protocol

C. Enter Number of Samples

D. Start the Run

V. Example of genomic DNA obtained from whole blood:

Agarose Gel Electrophoresis:

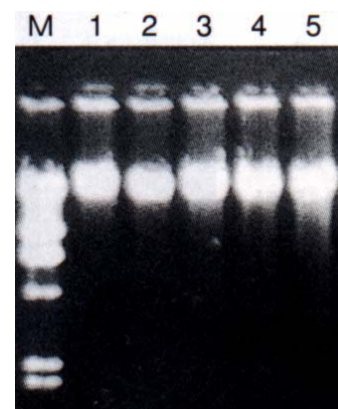
Sample: 3µl out of 250µl resuspended DNA

Gel: 0.5% TAE Gel

Condition: 80V x 2 hours

Lane M: High Molecular Weight DNA Marker (48.5 kb)

Lanes 1-5: DNA from 1ml whole blood.



VI. Extraction and Purification Process

Process Site	Purpose	System Process
1. Automated	Obtain nuclei pellet (includes mitochondria)	Add Buffer FG1*, centrifuge and discard supernatant.
2. Automated	Wash nuclei pellet	Add Buffer FG3*, centrifuge and discard supernatant. Repeat.
3. Automated	Digest/lyse nuclei	Add QIAGEN Protease and Buffer FG2*. Mix, incubate and mix again.
4. Automated	Precipitate DNA	Add R4** or Isopropanol to DNA recovery tube. Transfer supernatant of sample tube into DNA recovery tube, mix, centrifuge and discard the supernatant.
5. Automated	Wash DNA	Add R5** or 70% Ethanol, mix, centrifuge and discard the supernatant.
6. Automated	Resuspend DNA***	Add reagent FG3*, mix, incubate, mix and centrifuge.

* Buffers FG1, FG2 FG3 and QIAGEN Protease are available from AutoGen as AGFSTAR FlexiGene Reagent Kit (640) Catalog #AGKT-FG-640.

** Reagents R4 and R5 are available from AutoGen, Inc. as AutoGenFlex Blood DNA Finishing Kit, Catalog #AGFXPW.

*** Samples can be vortexed after initial incubation if required.

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