Genesee Scientific
A Life Science Company

Product Manual

GENE CHOICE
A Genesee Scientific Brand

PCR Purification Kit

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Introduction

The Gene Choice® PCR Purification Kit is an innovative system that radically simplifies the purification of nucleic acids from PCR and other impure reactions. The key to this system is the matrix that specifically, but reversibly, binds DNA or RNA under optimized conditions allowing proteins and other contaminants to be removed. Nucleic acids are easily eluted with deionized water or a low salt buffer.

Binding Capacity:
Each Micro DNA Column can bind ~10 µg of DNA.

Kit Contents

Product	SMP96-302	96-302	96-302B
Purifications	5	50	200
Micro DNA Columns	5	50	200
2 ml Collection Tubes	5	50	200
GC1 Buffer	5 ml	30 ml	120 ml
DNA Wash Buffer	2.5 ml	25 ml	3 x 25 ml
Elution Buffer	2 ml	30 ml	30 ml
User Manual	✓	✓	✓

Preparing Reagents

Dilute DNA Wash Buffer with 100% ethanol as follows and store at room temperature.

Kit	100% Ethanol to be Added
SMP96-302	10 ml
96-302	100 ml
96-302B	100 ml per bottle

Storage and Stability

All of the kit components are guaranteed for at least 12 months from the date of purchase when stored at room temperature. During shipment or storage in cool ambient conditions, precipitates may form in CP Buffer. Dissolve such deposits by warming the solution at 37°C and gently shaking.

Guidelines for Vacuum Manifold

The following is required for use with the Vacuum/Spin Protocol:

A) **Vacuum Manifold**
Compatible Vacuum Manifolds: Qiagen QIAvac24, Sigma AldrichVM20, Promega Vacman®, or manifold with standard Luer connector

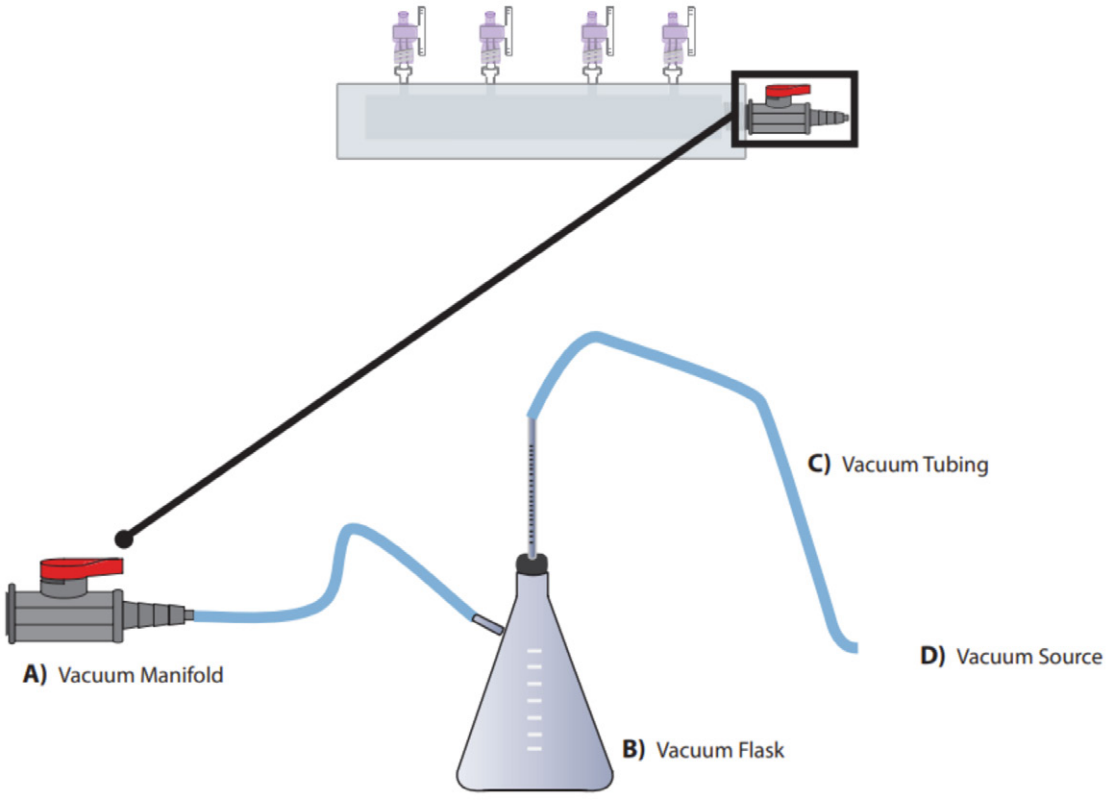
B) **Vacuum Flask**

C) **Vacuum Tubing**

D) **Vacuum Source** (review tables below for pressure settings)

Conversion from millibars:	Multiply by:
Millimeters of mercury (mmHg)	0.75
Kilopascals (kPa)	0.1
Inches of mercury (inch Hg)	0.0295
Torrs (Torr)	0.75
Atmospheres (atmos)	0.000987
Pounds Per Square Inch (psi)	0.0145

Vacuum Setup:



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Centrifugation Protocol

Materials and Equipment to be Supplied by User:

- Microcentrifuge capable of at least 13,000 x *g*
- 100% ethanol
- 1.5 ml microcentrifuge tubes
- Vortexer
- **Optional:** 3M NaOH
- **Optional:** Sterile deionized water or TE Buffer
- For fragments < 200 bp: 100% isopropanol

Before Starting:

- Prepare DNA Wash Buffer according to "Preparing Reagents" section

1. Perform agarose gel/ethidium bromide electrophoresis to analyze PCR product

2. Determine the volume of your PCR reaction

3. Transfer the sample into a clean 1.5 ml microcentrifuge tube

4. Add 5 volumes GC1 Buffer. For fragments < 200 bp, add 5 volume GC1 Buffer and 0.4 volumes 100% isopropanol
Note: Volume refers to the size of your PCR reaction. For example, if your PCR reaction is 50 µL, you would use 250 µL GC1 Buffer. If fragments are less than 200 bp, then add 250 µL GC1 Buffer and 20 µL 100% isopropanol.

5. Vortex to mix thoroughly. Briefly centrifuge to collect any drops from the inside of the lid

6. Insert a Micro DNA Column into a 2 ml Collection Tube (provided)

Optional Protocol for Column Equilibration:

- Add 100 µL 3M NaOH to the Micro DNA Column
- Centrifuge at 10,000 x *g* for 30 seconds
- Add 500 µL sterile deionized water to the Micro DNA Column
- Centrifuge at 10,000 x *g* for 30 seconds
- Discard the filtrate and reuse the collection tube

7. Transfer the sample from **Step 5** to the Micro DNA Column

8. Centrifuge at maximum speed (≥13,000 x *g*) for 1 minute at room temperature

9. Discard the filtrate and reuse collection tube

10. Add 700 µL DNA Wash Buffer
Note: DNA Wash Buffer must be diluted with 100% ethanol before use. Please see the "Preparing Reagents" section for instructions.

11. Centrifuge at maximum speed for 1 minute

12. Discard the filtrate and reuse collection tube

13. Repeat **steps 10-12** for a second DNA Wash Buffer wash step

14. Centrifuge the empty Micro DNA Column at maximum speed for 2 minutes to dry the column
Note: This step is critical for removal of trace ethanol that may interfere with downstream applications.

15. Transfer the Micro DNA Column into a clean 1.5 ml

16. Add 10-20 µL Elution Buffer, TE Buffer, or sterile deionized water directly to the center of column matrix

17. Let sit at room temperature for 2 minutes

18. Centrifuge at maximum speed for 1 minute
Note: This represents approximately 80-90% of bound DNA. An optional second elution will yield any residual DNA, though at a lower concentration.

19. Store DNA at -20°C

Vacuum Protocol

Materials and Equipment to be Supplied by User:

- Vacuum Manifold
- Microcentrifuge capable of at least 13,000 x *g*
- Nuclease-free 1.5 ml microcentrifuge tubes
- 100% ethanol
- Optional: 3M NaOH
- Optional: Sterile deionized water or TE Buffer
- For fragments < 200 bp: 100% isopropanol

Before Starting:

- Prepare DNA Wash Buffer according to the "Preparing Reagents" section

1. Perform agarose gel/ethidium bromide electrophoresis to analyze PCR product

2. Determine the volume of your PCR reaction

3. Transfer the sample into a clean 1.5 ml microcentrifuge tube

4. Add 5 volumes GC1 Buffer. For fragments < 200 bp, add 5 volume GC1 Buffer and 0.4 volumes 100% isopropanol
Note: Volume refers to the size of your PCR reaction. For example, if your PCR reaction is 50 µL, you would use 250µL GC1 Buffer. If fragments are less than 200 bp, then add 250 µL GC1 Buffer and 20 µL 100% isopropanol.

5. Vortex to mix thoroughly. Briefly centrifuge to collect any drops from the inside of the lid

6. Prepare the vacuum manifold according to manufacturer's instructions and connect the Micro DNA Column to the manifold

Optional Protocol for Column Equilibration:

- Add 100 µL 3M NaOH to the Micro DNA Column
- Switch on vacuum source to draw the buffer through the column
- Turn off the vacuum
- Add 500 µL sterile deionized water to the Micro DNA Column
- Switch on vacuum source to draw the water through the column
- Turn off the vacuum

7. Transfer the entire sample from **Step 5** to the Micro DNA Column

8. Switch on vacuum source to draw the sample through the column

9. Turn off the vacuum

10. Add 700 µl DNA Wash Buffer
Note: DNA Wash Buffer must be diluted with 100% ethanol before use. Please see the "Preparing Reagents" section for instructions.

11. Switch on vacuum source to draw the DNA Wash Buffer through the column

12. Turn off the vacuum

13. Repeat **steps 10-12** for a second DNA Wash Buffer wash step

14. Transfer the Micro DNA Column into a 2 ml Collection Tube (provided)

15. Centrifuge the empty Micro DNA Column at maximum speed for 2 minutes to dry the column
Note: This step is critical for removal of trace ethanol that may interfere with downstream applications.

16. Transfer the Micro DNA Column into a clean 1.5 ml microcentrifuge tube (not provided)

17. Add 10-20 µl Elution Buffer, TE Buffer, or sterile deionized water directly to the center of column matrix

18. Let sit at room temperature for 2 minutes

19. Centrifuge at maximum speed for 1 minute
Note: This represents approximately 80-90% of bound DNA. An optional second elution will yield any residual DNA, though at a lower concentration.

20. Store DNA at -20°C

Troubleshooting Guide

Please use this guide to troubleshoot any problems that may arise. For further assistance, **please contact the technical support staff**.

Possible Problems and Suggestions

Problem	Cause	Solution
Low DNA Yields	• Not enough GC1 Buffer added to sample	• Not enough GC1 Buffer added to sample
	• pH of the sample mixture is too high	• pH of the sample mixture is too high
No DNA Eluted	• DNA Wash Buffer was not diluted with ethanol	• Prepare DNA Wash Buffer as instructed

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