GeneDia

MagPure Cell-Free DNA Kit

GENEDIA™ **life Science Co**.

Product # EK1050R

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Introduction

The **GENEDIA™ MagPure Cell-Free DNA Kit** provides a rapid method for the isolation and purification of genomic DNA from Cell-Free samples. The **GENEDIA™ MagPure Cell-Free DNA Kit** is developed for scalable, rapid purification of high-quality DNA from serum samples. DNA purified with this Buffer can be used in a broad range of molecular biology downstream applications, such as sequencing, genotyping, and qPCR. This protocol guides you through manual isolations using a plate format. (**Magnetic** beads)

Buffer Specifications

GENEDIA™ MagPure Cell-Free DNA Kit is based on the use of magnetic beads that bind DNA under optimized binding conditions. **Two step Buffer** is added to the sample, mixed and incubated to bind the cf-DNA to nanoparticle, and the resulting solution is placed on the magnetic separation rack (**GENEDIA™ Magbead separation rack)**. Only the DNA will bind to the magnetic beads. The bound DNA is then washed in one step with ETOH 80% to remove any remaining impurities, and the purified total DNA is eluted with the PCR grade ddH20. The purified DNA can be used in a number of downstream applications.

Buffer Components

Components	labelling	Product # EK1050R (50 preps)
Lysis Buffer	Cf-DL	1 ml
GENEDIA™ MagPure Cell-Free DNA Isolation Buffer	Cf-Mag	15 ml
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Storage Conditions

All components of the **GENEDIA™ MagPure Cell-Free DNA Kit** should be stored at 25 °C and are stable for 1 year.

Recommended Equipment and Reagents

- 58-80 °C incubator
- Sampler in 100 to 1000 Microliter size
- Sampler tips
- 1.5 mL microcentrifuge tubes
- Vortex
- A magnetic separation racks. We recommend using the **GENEDIA™ Magbead separation rack** which is precisely compatible with the test procedure.
- Ethanol 80%

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Precautions and Disclaimers

- Prior to using the protocol, Genetic ID recommends that care be taken in homogenizing the sample in a manner that minimizes the risk of inadvertently contaminating the sample. Contamination can occur using improperly cleaned equipment or using poor laboratory practices during homogenization, weighing and labeling of the subsample.
- Perform all steps at room temperature (20–25°C) unless otherwise noted.

Sampling and Extraction Procedure

A. Sample preparation

The **GENEDIA™ MagPure Cell-Free DNA Kit** is compatible with serum/plasma samples separated from peripheral blood.

Note: *Sampling should be done in special tubes for cell free DNA (NOT provided). The unique preservative limits the release of genomic DNA, allowing isolation of high-quality cell-free DNA. Cell-Free DNA BCT has also been demonstrated to minimize the degradation of circulating tumour cells (CTCs). By limiting cell lysis, the specialized chemistry provides sample integrity during storage, shipping and handling of blood samples.*

B. Extraction procedure

1) Add **20 µl cf-DL** to **400 µl serum or plasma** sample Mix by vortex.
2) Incubate at **70°C** for **20 minutes**.
3) Centrifuge the for **15 minutes** at **13200 rpm**.
4) Transfer upper phase to new 1.5 microtube.
5) Add **300 µL** of **GENEDIA™ MagPureCell-Free DNA Buffer (Cf-Mag)** (shake vigorously before use, Shake the storage bottle well or place it on a vortex for a short length of time. Premixing magnetic beads with the binding buffer allows easier homogenous distribution of the beads to individual samples) and close the cap.
6) Incubate at room temperature for **15 minutes**. Up and down periodically.
7) Place the microtube in the **GENEDIA™Magbead separation rack** for **3 minutes**.
8) Keeping the microtube in the magnetic rack, carefully open the microtube cap and slowly aspirate and discard the leftover liquid. **Do not disturb the adsorbate**.
9) Add **1000µL**of **ETOH 80%.** Vortex for **30 seconds**.
10) Place the microtube in the **GENEDIA™Magbead separation rack** for **3 minutes**.
11) Keeping the microtube in the magnetic rack, carefully open the microtube cap and slowly aspirate and discard the leftover liquid. Do not disturb the adsorbate.
12) Add **100µL**of **PCR grade ddH20.**
13) Incubate for **5 minutes** at **90°C**. Vortex periodically.
14) Place the microtube in the **GENEDIA™Magbead separation** rack for **5 minutes**.
15) Transfer the liquid (including cfDNA) carefully into a new microtube.

C. Storage of DNA

The purified DNA is ready for immediate use. For short term storage, keep the extracted cfDNA in refrigerator at 2-8°C and for long term storage keep it in fridge at -20 to -70 °C.

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Troubleshooting Guide

Problem	Possible Cause	Solution and Explanation
Low DNA yield	**Very low load of cfDNA**	Increase the volume of the starting sample (up to 5 ml).
	Incomplete elution	Larger elution volumes and longer incubation times can increase yield.
	Reagents added incorrectly	Make sure that buffers have been reconstituted correctly, and that reagents have been added in the correct order.
	Incomplete elution during preparation	Larger elution volumes and longer incubation times can sometimes increase yield. Multiple rounds of elution can also be performed.
Low DNA performance	**Nucleic acids degraded**	Samples should be processed immediately. If necessary, add DNase inhibitor to the sample. Create a nuclease-free environment and ensure that no nucleases are present. Use suitable tips and buffer reservoirs. Check that all buffers have been prepared and stored correctly.

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