

NucleoGene Plant/Food DNA Extraction Kit

Instructions For Use

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REF

For isolation and purification of Deoxyribonucleic acid from Plant/Food samples.
For research implementation only.
Not suitable for diagnostic use.
For professional use only.

NGE021– 50 Tests
100 Tests
250 Tests

Kit Contents

	Supplied Material	50 Test	100 Test	250 Test
1.	Proteinase K*	1 vial	2 vial	5 vial
2.	Proteinase K Buffer	1 ml	2 ml	5 ml
3.	Lysis Buffer†	50 ml	100 ml	250 ml
4.	Wash Buffer I*	13 ml	26 ml	65 ml
5.	Wash Buffer II*	7ml	14 ml	35 ml
6.	Binding Buffer	22 ml	44 ml	110 ml
7.	Elution Buffer	11mL	22 ml	55 ml
8.	Spin Column	50	100	250
9.	Collection Tubes	50	100	250
10.	User's Guide	1	1	1

*Look at "Notes Before Starting" for preparation of Proteinase K and Wash Buffer.

†Precipitation may occur during storage. Heat to dissolve the precipitate formed before use.

Storage Conditions & Robustness

Kit components should be stored at room temperature (15-25°C) excluding Proteinase K. Do not expose Kit to direct sunlight. Proteinase K should be stored at -20 °C. When used under the second conditions, the kit can be stored for 12 months without any performance loss.

Intended Use

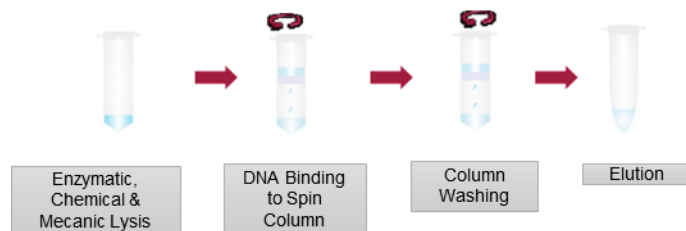
NucleoGene Plant/Food DNA Extraction Kit includes spin column and special lysis buffer system for effectively DNA extraction from Plant/Food samples (cereal, flour, soybean and soy milk, chocolate, chips, cereal dry, lecithin, oil, etc.). Specifically designed for DNA isolation and purification. Suitable for downstream applications such as purified DNA, PCR, Real Time Qualitative and Quantitative PCR (qPCR) and microarray. NucleoGene Plant/Food DNA Extraction Kit is designed for research use only. It is not for diagnosis and treatment of disease.

Product Usage Limits

- NucleoGene Plant/Food DNA Extraction Kit is for purification of nucleic acids of Plant/Food samples. It cannot be used for clinical purposes. Whether the kit is suitable for the user's own experimental design is up to the user's discretion.
- In particular, the ability of this kit to isolate DNA from Plant/Food samples has been confirmed.
- The collection, transport and storage of the sample to be used are at least important as purification and are sensitive stages that may affect the results.
- It is designed for professional use by educated personnel only.
- Kits or any kit components with different lot numbers should not be used together.

Introduction

NucleoGene Plant/Food DNA Extraction Kit is improved for cell DNA extraction effectively from Plant/Food samples (plants, cereal, flour, soybean and soy milk, chocolate, chips, cereal dry, lecithin, oil, etc.) to be a quick and useful method. Working principle of Kit depends on binding of DNA with its especially improved solutions to spin columns. The superior performance of the kit is due to its ability to lyse the cell walls of further-processed Plant/Food samples, as well as its ability to remove PCR inhibitors present in the sample with solutions specially improved for Plant/Food samples. While wash buffer optimized specifically is improved for purified nucleic acids especially from protein and other inhibitors, the Lysis Buffer solution is improved for lysing the cell wall and remove the inhibitors from the sample. With the complete the isolation process, DNA is obtained at high efficiency and purity.



NucleoGene Plant/Food DNA Extraction Kit is optimized for use in 50-200 mg Plant/Food sample. Purified DNA with completed isolation should be use immediately or a freezer at -20 °C should be preferred for long-term storage.

Warnings & Precautions

- All clinical specimens and residues and debris arising therefrom must be treated as potential infectious agents and disposed of accordingly.
- All samples should be prepared in Biosafety Level 1 and 2 or Class II Biosafety Cabinets.
- All surfaces should be cleaned daily with a disposable paper towel or napkin by using freshly prepared Sodium Hypochlorite %20 Solution.
- Do not neglect to use laboratory safety consumable materials such as disposable gloves, goggles, visor, disposable cuffs, and disposable mask.
- If any of the kit components come into contact with your skin, wash it with plenty of water immediately. In case any of the kit components come into contact with your mucous membranes such as your eyes or mouth, wash the contact part with plenty of water again; and go to the doctor. If possible, prefer filtered pipette tips.
- Do not mix the kit solutions or waste appeared during isolation with bleach.
- Keep Kit out of contamination sources such as DNA and RNA, especially amplified nucleic acid.
- Do not mix solutions with different lot numbers or do not use or combine products from other companies.
- For more information, please apply the Material Safety Data Sheet(MSDS), which you can request from www.nucleogene.com.

Other Required Materials

- Thermal Block/water bath
- Ethanol (%96-100)
- Microcentrifuge
- Vortex mixer
- Microcentrifuge tubes (1.5 ml)
- Mikrocentrifuge tubes (2.0 ml)
- Micropipette set and sterile filtered micropipette tips
- Spatula

Notes Before Starting

If the kit is kept in a non-air-conditioned environment, Lysis buffer solution can precipitate at room temperature in cold weather. In such a case to dissolve the precipitate, the Lysis buffer solution is held at 50°C in a water bath until dissolving completely. Be sure of all solutions are at the room temperature before and after protocol implementation, if they are not, allow them to reach room temperature.

-Add 13 mL of absolute or 96 % ethanol in Wash I Buffer bottle for 50 tests.

-Add 26 mL of absolute or 96 % ethanol in Wash I Buffer bottle for 100 tests.

-Add 65 mL of absolute or 96 % ethanol in Wash I Buffer bottle for 250 tests.

-Add 28 mL of absolute or 96 % ethanol in Wash II Buffer bottle for 50 tests.

-Add 56 mL of absolute or 96 % ethanol in Wash II Buffer bottle for 100 tests.

-Add 140 mL of absolute or 96 % ethanol in Wash II Buffer bottle for 250 tests.

Then, do not forget shaking with your hand or with vortex at least 10 seconds.

Proteinase K is supplied as lyophilizing. Before starting to use the kit, all lyophilized Proteinase K should be mixed with Proteinase K buffer (Each lyophilized Proteinase K tube should be mixed with 1 ml of Proteinase K buffer).

Protocol

Control the reagents

Before starting to DNA isolation, control reagents that are free of leakage. To mix the solutions, shake the reagents slightly. If reagents precipitate (specially Lysis solution), they dissolve at 50 ° C by heating.

Plant/Food DNA Isolation

1. Transfer (200 mg) of Plant/Food sample to a 2 mL tube, add the appropriate amounts of Lysis Buffer to samples indicated in the table and then vortex the tube for mixing and then add 20 µL of Proteinase K to the tube. Incubate the tube in thermal shaker block at 60 °C for 30-180 minutes (vortex the tubes for 5 seconds every 5 minutes when incubated in the fixed block). If you want an RNA-free result, you can add 5 µL RNase (10mg/mL) to the tube.

Plant/Food Type	Sample	Amount of Lysis Buffer to be used
Plant	All plant tissues	0,8 ml
Meat Products	Jambon, salami, sausage, fermented sausage, carcass etc	0,8 ml
Canned	Fish, vegetables, corn etc.	0,8 ml
Cereals	Cereal dry, chips, cake, biscuit, pasta etc.	1 ml
Milk products, Chocolate, and chocolate products	Yoghurt, cheese, chocolate, wafer etc.	0,8 ml
Flour and flour products	Flour, corn flour, soybean flour, ready cake mixtures	1 ml
Prepared Plant/Foodstuffs	Soup, toffees including gelatin, aroma etc.	1 ml

2. After incubation, centrifuge the tube at 11000xg for 10 minutes. Then transfer the lysate (400 µL) to a new 2 mL of the tube, add 400 µL Binding Buffer (**shake vigorously just before use**) and mix (vortex for 15 seconds or pipetting). Subsequently, transfer all liquid to spin column placed into the collection tube.

3. Centrifuge spin columns at 11000xg for 2 minutes.

4. Place the spin columns to a new collection tube, add 500 µL Wash I Buffer and centrifuge at 11000 x g for 1 minute.

5. Place the spin columns to a new collection tube, add 650 µL Wash II Buffer and centrifuge at 11000 x g for 1 minute. Repeat this step.

6. Place the spin columns to a new collection tube and centrifuge at maximum rate for 2 minutes to remove Ethanol residue.

7. Place the spin columns into 1.5 ml microcentrifuge tubes. Transfer 100-200 µL of Elution Buffer pre-heated to 60 °C to spin column and incubate for 1 minute at room temperature.

9. After incubation, centrifuge spin columns at 11000 x g for 1 minute. Throw the spin columns and close the cap of tubes.

10. Liquid in the tube contains DNA and ready for use. DNA is kept at -20 °C for long time storage.



Troubleshooting

Issue	reason	solution
Clogged Column	Overuse sample	If more than 200 mg of sample is used, increase the Proteinase K, Solution volumes by the same rate. Pass the lysate through a column.
	Incompleted lysis	Be sure for mixing well of sample and lysis solution.
Low DNA amount	Wrong washing	Verify that the wash solution concentrates are diluted with the specified volumes of ethanol. To prevent evaporation, close the caps of the bottles thoroughly.
	Weak elution	Reduce Elution volume.
	Clogged column	Lookup
Enzymatic reactions do not run with isolated DNA.	Low DNA concentration	Precipitate DNA with ethanol and then dissolved with low volume of Elution.
	High salt content of isolated DNA	Precipitate DNA with ethanol.
	Ethanol remaining from washing buffer	After washing stage, centrifuge the spin column to remove remaining ethanol.





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