

Instructions for Use

Life Science Kits & Assays



PME Food Enrichment Tool

1 Product specifications

Starting material	Liquid and soluble foods
Time of preparation	30 - 60 minutes

Enrichment of cell-free and cell-integrated DNA from liquid and instant foods for subsequent DNA isolation

2 Intended use

The content of DNA in liquid or instant foods is usually very low and the DNA is highly degraded. Because of these facts, the extraction of DNA from such foods is difficult. The PME Food DNA Enrichment Tool is based on a new technology, called: PME – Polymer Mediated Enrichment.

The procedure starts with capturing of cells and cell-free DNA from a large amount of liquid sample by a special polymer.

If necessary, the instant samples have to be dissolved in distilled water and/or proteins have to be digested with Proteinase K (see table below).

The next step is the centrifugation of captured DNA. Subsequently, the pellet containing DNA can be dissolved directly in a lysis buffer of kit used for DNA isolation.

ATTENTION!

Not all Lysis Solutions are suitable for dissolving of PME pellet. Use recommended DNA Isolation Kits from IST Innuscreen GmbH (see "Related Products").

Intended use

Kind of food	Amount	Dissolving (ddH ₂ O)	Proteinase K
Milk, liquid milk products (cream, shake etc.)	1 ml	no	no
Thick milk products (yoghurt, condensed milk, kefir)	1 g	300 µl	yes
Dry milk products (baby milk, dry milk)	0,6 g	1000 µl	yes
Juice, vinegar, wine	1 ml	no	no
Dry instant products without or with low amount of milk (3in1 coffee etc.)	0,6 g	1000 µl	no

ATTENTION!

The final volume of the starting material can be increased up to 10 ml. In this case use a 15 ml centrifugation tube instead of a 2.0 ml tube for the sample preparation. Follow the additional instructions (in italics bold letters) of the appropriate protocol.

In this case, the number of samples, which can be treated with this kit, is accordingly reduced.

The kit contains a Carrier RNA/DNA Mix. Addition of Carrier Mix is recommended if extreme low amount of DNA is expected. In this case the addition of Carrier Mix can increase the final yield. The artificial DNA inside of Carrier Mix can be used to evaluate nucleic acid extraction and to verify the absence of inhibitors during amplification.

The assay is intended for research use only.

The kit has been designed to be used for a wide range of different downstream DNA isolation methods (see "Related Products").

3 Product and order number

Name	Amount	Order-no.
PME Food DNA Enrichment Tool	50 rxn	845-IR-0009050





4 Storage conditions

Store lyophilized and dissolved Proteinase K and Enrichment Reagent VCR-1 at 4 °C to 8 °C. lyophilized and dissolved Carrier Mix have to be stored at -22 °C to -18 °C.

All other components of the Kit should be stored dry, at room temperature (15 °C to 30 °C).

When stored as recommended, the kit is stable until the expiration date printed on the label on the kit box.

5 Delivered components

Components		Σ 50
Proteinase K		For 1.5 ml working solution
Enrichment Reagent VCR-1		2 x 1.2 ml
Carrier Mix		For 1.25 ml working solution
Enrichment Reagent VCR-2		10 ml

Components not included in the kit

- ddH₂O for dissolving Proteinase K, Carrier Mix and samples
- 2 ml reaction tubes

6 Safety precautions

The assay shall only be handled by educated personal in a laboratory environment. The compliance with the specified procedure is absolutely mandatory when performing this assay.

Reagents should be stored in their original containers at the indicated temperatures. Do not replace individual components with those from different batches or test assays. Note the indicated expiration dates.

Do not eat, drink or smoke while performing the assay.

Wear protective clothing and safety gloves.

All samples and test materials should be handled and disposed of as infectious material, in accordance with regulatory requirements.

Reagent containers that have not come in contact with potentially infectious material may be disposed of along with ordinary laboratory waste.

7 DNA enrichment from food samples containing high amounts of proteins

Recommended steps before starting

- Heat thermal mixer or water bath at 50 °C for Proteinase K digestion.
- Ensure that Proteinase K and Carrier Mix have been prepared according to the instruction.
- Centrifugation steps should be carried out at room temperature.
- Avoid freezing and thawing of starting material.

1. Add an appropriate amount (see the table in “Intended use”) of sample into a 2.0 ml (**or 15 ml**) reaction tube. Dilute the sample if necessary.
2. Add 30 µl (**50µl**) Proteinase K and 10 µl (**100 µl**) of dissolved Carrier Mix, mix vigorously by pulsed vortexing for 10 seconds and incubate the sample at 50 °C for 30 minutes.

We recommend using a shaking platform (thermal mixer, water bath or another rocking platform) for a continuous shaking of the sample. No shaking will reduce the lysis efficiency.

3. Centrifuge the 2.0 ml reaction tube at 11,000 x g (~12,000 rpm) **or 15 ml reaction tube at 4,200 x g**

(~5,000 rpm) for 30 seconds to remove the non-dissolved parts. Transfer the supernatant into a new 2.0 ml **(15 ml)** reaction tube.

4. Open the reaction tube and add 30 μ l **(100 μ l)** of Enrichment Reagent VCR-1. Mix shortly by vortexing, add 150 μ l **(600 μ l)** of Enrichment Reagent VCR-2 to the tube, mix shortly by vortexing. Incubate at room temperature for 10 minutes.
5. Centrifuge at maximum speed ($>10,000 \times g / > 4,200 \times g$) for 10 minutes, open the tube and remove the supernatant carefully as much as possible. Possibly formed upper fraction (including fat) must also be removed.
6. After discarding the supernatant, the sample is ready for subsequent lysis step of chosen DNA isolation Kit (see "Related products").

8 DNA enrichment from food samples containing low amounts or no protein

Recommended steps before starting

- Ensure that Carrier Mix has been prepared according to the instruction.
 - Centrifugation steps should be carried out at room temperature.
 - Avoid freezing and thawing of starting material.
1. Add an appropriate amount (s. the table in "intended use") of sample into a 2.0 ml **(or 15 ml)** reaction tube. Dilute the sample if necessary. You can increase the sample amount
 2. Centrifuge the 2.0 ml reaction tube at $11,000 \times g$ ($\sim 12,000$ rpm) **or 15 ml reaction tube at $4,200 \times g$ ($\sim 5,000$ rpm)** for 30 seconds to remove the non-dissolved parts. Transfer the supernatant into a new 2.0 ml **(15 ml)** reaction tube.
 3. Open the reaction tube and add 10 μ l **(100 μ l)** of dissolved Carrier Mix and 30 μ l **(100 μ l)** of Enrichment Reagent VCR-1. Mix shortly by vortexing, add 150 μ l **(600 μ l)** of En-

richment Reagent VCR-2 to the tube, mix shortly by vortexing. Incubate at room temperature for 10 minutes.

4. Centrifuge at maximum speed ($>10,000 \times g / > 4,200 \times g$) for 10 minutes, open the tube and remove the supernatant carefully as much as possible. Possibly formed upper fraction (including fat) must also be removed.
5. After discarding the supernatant, the sample is ready for subsequent lysis step of chosen DNA isolation Kit (see "Related products").

9 Related products

Recommended DNA Extraction Kits	
Product	Order Number
innuPREP DNA Mini Kit 2.0	845-KS-1042...
blackPREP Rodent Tail DNA Kit	845-BP-0010...
Assay for Detection of Carrier DNA	
innuDETECT Internal Control DNA/RNA assay	845-ID-0008...
Some Assays for Food Detection	
innuDETECT Beef Assay	845-IDF-0020...
innuDETECT Horse Assay	845-IDF-0030...
innuDETECT Goat Assay	845-IDF-0040...
innuDETECT Sheep Assay	845-IDF-0050...
innuDETECT Fish Assay	845-IDF-0100...
innuDETECT Cheese Assay	845-IDF-0110...
innuDETECT Halal Assay	845-IDF-0120...
and much more...	

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