GENOMIC DNA EXTRACTION KIT FROM FFPE SAMPLES

General instructions:

To ensure proper use and handling, please, READ THE ENTIRE MANUAL BEFORE using the Kit.

Labelling the top of each vial upon arrival of the kit is highly recommended to avoid mistakes.

Caution: **PRB solution contains xylene, please handle with care in a fume extraction cupboard*

This DNA Kit has been designed for research use only from FFPE tissue sections.

The processing time after tissue digestion is approximately 90 minutes.

Kit contents

	20 extractions kit	50 extractions kit
PRB solution	22 ml	2x 26 ml
Solution A	7 ml	16 ml
Solution B	2.5 ml	6 ml
Solution C	0.85 ml	2.5 ml
Solution D	10 ml	22 ml
Solution E	12 ml	25.5 ml
Solution F	1 ml	3 ml
2 ml tubes	20	50
1.5 tubes	20	50

Equipment and materials required but not supplied

The following equipment and materials are required:

Pipets and pipet tips (pipet tips with aerosol barriers are strongly recomended to prevent cross-contamination)

Disposable gloves

Heating block for lysis of samples at 56°C and incubation at 90°C

Microcentrifuge

Vortexer

Fume extraction cupboard

Technical considerations

This Kit was specially designed with the aim of obtaining, in a reproducible manner, genomic DNA for subsequent uses in genetic diagnostic procedures.

Purified genomic DNA has been successfully tested specifically, both in quality and in quantity. However, DNA from FFPE samples is usually of lower molecular weight than DNA from fresh samples. DNA fragmentation depends on the age and type of the sample and the conditions used for fixation.

Identification of the substance/mixture

PRB SOLUTION (Paraffin Remove Buffer)

Contains Xylene. Xylene is a hazardous chemical that must be used in a fume hood, and the resulting waste handled appropriately.

1. HAZARDS IDENTIFICATION

a. Classification of the substance or mixture

- Classification according to Regulation (EC) No 1272/2008

Flammable liquids (Category 3)
Acute toxicity, Inhalation (Category 4)
Acute toxicity, Dermal (Category 4)
Skin irritation (Category 2)

- Classification according to EU Directives 67/548/EEC or 1999/45/EC

Flammable. Harmful by inhalation and in contact with skin. Irritating to skin. Flammable. Harmful by inhalation and in contact with skin. Irritating to skin.

b. Label elements

2. HANDLING AND STORAGE

a. Precautions for safe handling

Avoid contact with skin and eyes. Avoid inhalation of vapour or mist.
Keep away from sources of ignition - No smoking. Take measures to prevent the build up of electrostatic charge.

b. Conditions for safe storage, including any incompatibilities

Store in cool place. Keep container tightly closed in a dry and well-ventilated place. Containers which are opened must be carefully resealed and kept upright to prevent leakage.

3. ACCIDENTAL RELEASE MEASURES

a. Personal precautions, protective equipment and emergency procedures

Use personal protective equipment. Avoid breathing vapors, mist or gas. Ensure adequate ventilation.
Remove all sources of ignition. Beware of vapours accumulating to form explosive concentrations.
Vapours can accumulate in low areas.

b. Environmental precautions

Prevent further leakage or spillage if safe to do so. Do not let product enter drains. Discharge into the environment must be avoided.

c. Methods and materials for containment and cleaning up

Contain spillage, and then collect with an electrically protected vacuum cleaner or by wet-brushing and place in container for disposal according to local regulations.

SOLUTION A

Non Hazardous

SOLUTION B

Non Hazardous **SOLUTION C:** Contains sodium acetate solution

1. HAZARDS IDENTIFICATION

a. Classification of the substance or mixture

- Classification according to Regulation (EC) No 1272/2008

Eye irritation (Category 2)
Not a hazardous substance or mixture according to EC-directives 67/548/EEC or 1999/45/EC.

b. Label elements

2. HANDLING

a. Precautions for safe handling

Avoid contact with skin and eyes. Avoid inhalation of vapour or mist.
Normal measures for preventive fire protection.

3. ACCIDENTAL RELEASE MEASURES

a. Personal precautions, protective equipment and emergency procedures

Use personal protective equipment. Avoid breathing vapors, mist or gas.
Ensure adequate ventilation.

b. Environmental precautions

Do not let product enter drains.

c. Methods and materials for containment and cleaning up

Soak up with inert absorbent material and dispose of as hazardous waste. Keep in suitable, closed containers for disposal.

SOLUTION D: Contains 2-Propanol

1. HAZARDS IDENTIFICATION

a. Classification of the substance or mixture

- Classification according to Regulation (EC) No 1272/2008

Flammable liquids (Category 2)
Eye irritation (Category 2)
Specific target organ toxicity - single exposure (Category 3)

- Classification according to EU Directives 67/548/EEC or 1999/45/EC

Highly flammable. Irritating to eyes. Vapours may cause drowsiness and dizziness.

b. Label elements

2. HANDLING

a. Precautions for safe handling

Avoid contact with skin and eyes. Avoid inhalation of vapour or mist.
Keep away from sources of ignition - No smoking.Take measures to prevent the build up of electrostatic charge.

b. Skin protection

Handle with gloves. Gloves must be inspected prior to use.
Use proper glove removal technique (without touching glove's outer surface) to avoid skin contact with this product. Dispose of contaminated gloves after use in accordance with applicable laws and good laboratory practices. Wash and dry hands.

3. ACCIDENTAL RELEASE MEASURES

a. Personal precautions, protective equipment and emergency procedures

Use personal protective equipment. Avoid breathing vapors, mist or gas.
Ensure adequate ventilation. Remove all sources of ignition.
Evacuate personnel to safe areas. Beware of vapours accumulating to form explosive concentrations. Vapours can accumulate in low areas.

b. Environmental precautions

Prevent further leakage or spillage if safe to do so. Do not let product enter drains.

SOLUTION E

Non Hazardous

SOLUTION F

Non Hazardous

Storage

Keep container tightly closed in a well-ventilated place.

All buffers can be stored at room temperature (15-25°C). If temperature exceeds 25°C, is recommended to store, at least solution A and C, in a cool place (2-8°C). White precipitates can be formed in solution A when stored in a cool place.

Remember, if stored at (2-8°C) solutions should be homogenized and equilibrated to room temperature before use (especially solution A to dissolve white precipitates formed).

All buffers are stable for at least 1 year when stored at room temperature (15-25°C) but only until the kit expiration date (see box label). If stored at 4°C the kit is stable for more than 1 year and quality does not decrease.

Procedure recommendations

- Samples

The extraction procedure can be performed from freshly cut sections of FFPE tissue. 5-10 sections of up to 10 µm can be used in one preparation.

- Solutions
Gently homogenize every solution before use, especially solutions A and C if stored in a cool place.
- Dry
It is recommended not overdrying the final pellet in order to avoid a difficult DNA solubilization. Add F solution when the pellet starts drying.

DNA Quantity and quality
Concentration
Fluorometric based method can be used in order to get accurate and reliable concentration readings, particularly when no RNase treatment has been done, as traces of RNA may over-quantify the sample concentration.

Ratio 260/230 and 260/280
DNA quality can be determined based on 260/230 and 260/280 ratios, however, with small DNA amount or RNA traces, ratios may change without affecting DNA quality (specially 260/230 ratio).