XIT™ Genomic DNA from Plant Tissue

For the isolation of genomic DNA from fresh or frozen tissue

INTRODUCTION

The *XIT*™ Genomic DNA from Plant Tissue kit is designed for the isolation of genomic DNA from fresh or frozen plant tissue. The *XIT*™ kit uses the principle of cell lysis, protein digestion and precipitation and finally DNA precipitation to isolate high quality genomic DNA.

XIT™ Genomic DNA from Plant Tissue kit protocol is designed to use 10-100mg plant tissue, however the protocol can be easily adapted for larger tissue sample sizes. The purified DNA has an A_{260}/A_{280} ratio between 1.8-2.0 and has yields ranging between 1-5µg/mg depending on plant species.

ITEM(S) SUPPLIED	Cat # 786-297 *For 2.5g tissue*	Cat # 786-298 *For 25g tissue*
XIT™ Lysis Buffer	10ml	100ml
LongLife™ Proteinase K	0.5ml	12.5ml
XIT™ Protein Precipitation Buffer	2.5ml	25ml
TE Buffer	1.5ml	20ml
LongLife™ RNase	0.5ml	0.5ml

STORAGE CONDITIONS

The kit is shipped at ambient temperature. Upon arrival, store the *LongLife*™ Proteinase K and *LongLife*™ RNase at -20°C, store all other kit components at room temperature. The kit components are stable for 1 year, if stored properly.

ITEMS NEEDED BUT NOT SUPPLIED

Isopropanol, 70% ethanol

PREPARATION BEFORE USE

1. Preheat a waterbath or heating block to 55°C.
2. Equilibrate TE Buffer to 50-60°C.

PROTOCOL

1. For optimal yield, freeze 10-100mg plant tissue in liquid nitrogen and quickly grind in liquid nitrogen with a pestle and mortar. Keep the tissue on ice at all times.

   NOTE: If liquid nitrogen is not available, freeze the tissue and rapidly grind or homogenize on ice.

   NOTE: For efficient grinding, we recommend G-Biosciences' EZ-Grind™ *(Cat. # 786-139), a high efficient grinding resin with matching pestle and tubes.*
2. Transfer the ground or homogenized tissue to a 1.5ml microfuge tube and add 400µl *XIT*™ Lysis Buffer. If large clumps are visible grind the tissue further in the presence of the lysis buffer.
3. Add 20µl *LongLife*™ Proteinase K to the tube and mix by inverting the tube 10-20 times. Incubate at 55°C overnight for maximal yield. Invert the tube periodically during the incubation.
4. Add 90µl *XIT*™ Protein Precipitation Buffer to the sample and mix by inverting the tube 10-20 times.
5. Centrifuge at 16,000g for 5 minutes. Carefully, transfer the supernatant to a fresh tube.

NOTE: The supernatant should be clear. If not, repeat the centrifugation.

6. Add 400µl isopropanol to the supernatant and mix by gently inverting the sample at least 20-25 times.
7. Centrifuge at 14,000rpm for 5 minutes.
8. Discard the supernatant and use a pipette to carefully remove remaining liquid without disturbing the pellet.
9. Add 200µl 70% ethanol and invert the tube twice to wash the pellet.
10. Centrifuge at 14,000rpm for 10 minutes.
11. Discard the supernatant and drain the tube on a piece of clean absorbent paper. Allow to air dry for 15 minutes.
12. Add 50µl prewarmed TE buffer and 1µl *LongLife*™ RNase to remove the RNA (if required).
13. Rehydrate the genomic DNA by incubating at 55-65°C for one hour, followed by an overnight incubation at room temperature to ensure complete genomic DNA hydration.
14. Store DNA at 4°C, for long term storage store at -20 or -80°C.

RELATED PRODUCTS

1. ***EZ-Grind™ (Cat # 786-139)****: A highly efficient grinding resin that is pre-aliquoted into 1.5ml grinding tubes and is supplied with matching pestles.*
2. ***Pestle & Tubes (Cat. # 786-138P):*** *DNase/RNase free microfuge tubes (1.5ml) and matching pestles for the grinding of small samples and isolation of nuclei.*
3. ***Molecular Grinding Resin™ (Cat # 786-138):*** *For grinding of small samples. High tensile micro particles that do not bind nucleic acids, allowing most samples to be processed by hand using inexpensive micro centrifuge tube pestles or a mortar and pestle.*

NOTE: *For other related products, visit our web site at www.GBiosciences.com or contact us.*