Ver 2.0

HB1170

Handbook for

- PLANT SV MINI
- PLANT SV MIDI
- PLANT SV MAXI

exgene™

DNA PURIFICATION HANDBOOK

GeneAll

Customer & Technical Support

Do not hesitate to ask us any question.

We thank you for any comment or advice.

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This protocol handbook is included in :

GeneAll® Exgene™ Plant SV mini (117-101, 117-152)
GeneAll® Exgene™ Plant SV Midi (117-226, 117-201)
GeneAll® Exgene™ Plant SV MAXI (117-310, 117-326)

Visit www.geneall.com or www.geneall.co.kr for FAQ, QnA and more information.

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	Plant SV mini		Plant SV Midi	
Cat. No.	**117-101**	**117-152**	**117-226**	**117-201**
Size	**mini**	**mini**	**Midi**	**Midi**
No. of preparation	100	250	26	
GeneAll® SV column type G (with collection tube)	100	250	26	
EzSep™ filter column (with collection tube)	100	250	26	**117-226 X 4**
Buffer PL	100 ml	200 ml	100 ml	
Buffer PD	30 ml	90 ml	30 ml	
Buffer BD (concentrate) *	37 ml	51 ml x 2	37 ml	
Buffer CW (concentrate) * †	30 ml	40 ml x 2	50 ml	
Buffer AE **	60 ml	120 ml	60 ml	
RNase A (100 mg/ml)	0.48 ml	1.3 ml	0.48 ml	
Protocol Handbook	1	1	1	

	Plant SV MAXI	
Cat. No.	**117-310**	**117-326**
Size	**MAXI**	**MAXI**
No. of preparation	10	26
GeneAll® SV column type G (with collection tube)	10	26
EzSep™ filter column (with collection tube)	10	26
Buffer PL	100 ml	200 ml
Buffer PD	30 ml	90 ml
Buffer BD (concentrate) *	37 ml	51 ml x 2
Buffer CW (concentrate) * †	50 ml	50 ml x 3
Buffer AE **	60 ml	120 ml
RNase A (100 mg/ml)	0.48 ml	1.3 ml
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** Before using for the first time, add absolute ethanol (ACS grade or better) into buffer BD and CW as indicated on the bottle.*

† *Contains sodium azide as a preservative*

*** 10 mM TrisCl, pH 9.0, 0.5 mM EDTA*

Storage Conditions

All components of GeneAll® Exgene™ Plant SV kit should be stored at room temperature (15~25°C). RNase A is delivered under ambient conditions and can be stored at room temperature for 6 months without significant decrease in activity. But for prolonged conservation of activity, storing at -20~8°C is recommended.

During delivery or storage under cold ambient condition, a precipitate may be formed in buffer PL. Heat the bottle to dissolve completely before use. Using precipitated buffers will lead to poor DNA recovery. GeneAll® Exgene™ Plant SV kit series are guaranteed for 1 year.

Quality Control

All components of GeneAll® Exgene™ Plant SV kit are manufactured in strictly clean condition, and its degree of cleanness is monitored periodically. Restriction enzyme assay, PCR amplification assay and spectrophotometric assay as quality control are carried out from lot to lot thoroughly, and only the qualified is delivered.

Chemical Hazard

The buffers included in GeneAll® Exgene™ Plant SV kit contain irritant which is harmful when in contact with skin or eyes, or when inhaled or swallowed. Care should be taken during handling. Always wear gloves and eye protector, and follow standard safety precautions.

Buffer BD contains chaotropes. It can form highly reactive compounds when combined with bleach. Do NOT add bleach or acidic solutions directly to the sample-preparation waste.

Product Specifications

GeneAll® Exgene™ Plant SV	mini	Midi*	MAXI*
Sample amount	~100 mg wet	~400 mg wet	~1 g wet
Preparation time	<40 min	<1 hour	<1 hour
Maximum loading volume	750 ul	5 ml	15 ml
Binding capacity	50 ug	170 ug	400 ug
Typical yield	4-40 ug	10-150 ug	40-300 ug
Elution volume	30-400 ul	200-600 ul	0.4-2 ml

** GeneAll® Exgene™ Plant SV Midi/MAXI kit procedures require the centrifuge which has a swinging-bucket rotor and ability of 4,000~5,000 x g.*

GeneAll® Exgene™ Plant SV

Introduction

GeneAll® Exgene™ Plant SV kit provides a simple and easy method for the small, medium and large scale purification of total DNA from various plant tissues.
With EzSep™ filter and GeneAll® SV column type G, several plant metabolites are efficiently removed and the procedure can be done in just 40 minutes (mini), yielding a pure DNA suitable for various downstream applications without further manipulation. Up to 100 mg, 400 mg and 1,000 mg of plant tissue can be processed with GeneAll® Exgene™ Plant SV mini, Midi and MAXI, respectively. GeneAll® Exgene™ Plant SV procedure eliminates the need of organic solvent extraction and alcohol precipitation, allowing safe and fast preparation of many samples simultaneously. Purified total DNA can be directly applicable in conventional PCR, real time PCR, Southern blotting, SNP genotyping, RFLP, AFLP and RAPD.

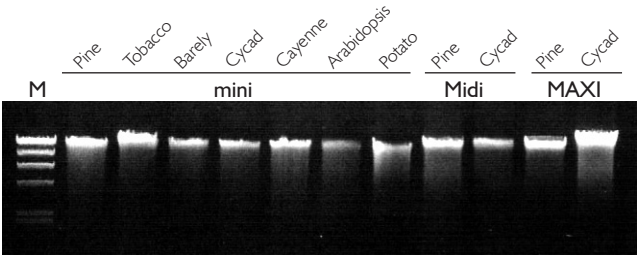


Fig 1. Genomic DNA prepared from various plant tissues using GeneAll® Exgene™ Plant SV series. Purified DNA was resolved on 0.7% agarose gel.

Scale	mini	Midi	MAXI
Sample weight	100 mg	400 mg	1,000 mg
Elution vol.	100 ul	400 ul	1,000 ul
Loaded vol.	5 ul	5 ul	10 ul

Source	DNA yield (ug)
Arabidopsis	2~5
Barely	4~10
Cayenne	4~18
Cycad	4~15
Maize	7~16
Pine	6~20
Potato	2~8
Soybean	3~15
Tobacco	7~25

Typical yield from various plant tissues (100 mg) with GeneAll® Exgene™ Plant SV mini kit *DNA yields vary depending on several factors; age, regions, genome size, stored conditions, and harvest or disruption methods of plant tissue. Midi procedures may yield usually DNA of 3~4 times to mini, and approximately 10 times with MAXI.*

General Considerations

Starting sample amount

There is an optimized sample amount for GeneAll® Exgene™ Plant SV kit procedures. For mini kit, 100 mg (wet weight) of starting sample material is optimized for the procedures. For dried or lyophilized tissue, it is 25 mg. If the size of starting sample is larger than the optimized, tissue lysis can not be performed efficiently, and this will bring about poor DNA recovery. For large amount of sample, GeneAll® Exgene™ Plant SV Midi/MAXI is available.

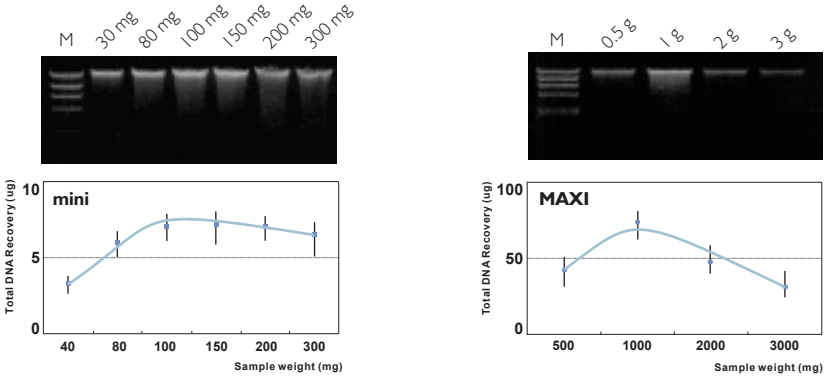


Fig 2. DNA Preparation from pine.
Use of an excessive starting sample may cause incomplete lysis of sample tissues and the shearing of DNA, resulting in low yield and poor quality of DNA. 2 ul out of 100 ul eluate was resolved on 0.8 % agarose gel. M : Lambda-HindIII

Sample preparation, pulverization and lysis

When purifying DNA from plants, harvest and pulverization of sample is the most important step for good result. Harvested plant sample or ground tissue powder should be stored under -70°C after frozen in liquid nitrogen for future use. Lyophilized tissue can be stored at room temperature. Fresh and young plant tissues would be best for high yield and good quality of DNA.

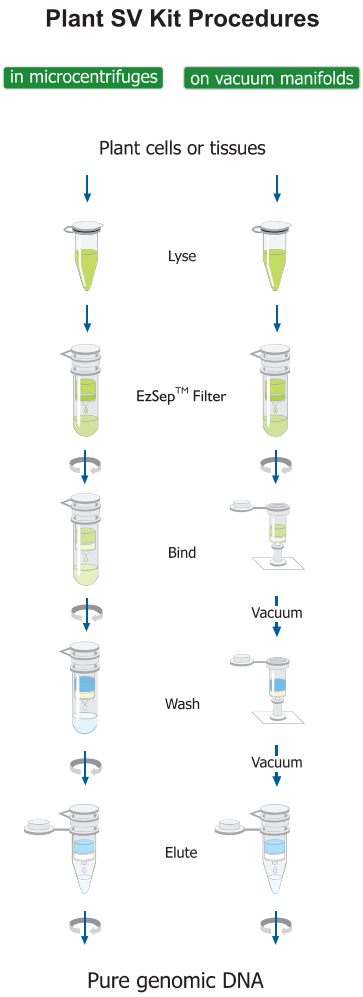
Before lysis, tissue sample should be disrupted completely for efficient lysis, and this step should be performed at low temperature (below 0°C) as quickly as possible for optimized result. Lyophilized tissue can be ground at ambient condition.
Mortar and pestle with liquid nitrogen is a typical and good method for grinding of sample. Rotor-stator homogenizer or bead-beater can be a good alternative. Complete and quick pulverization of sample tissue will guarantee the optimized result, while incomplete ground sample or the sample thawed by delayed or poor handling may result in low yields and degraded DNA.

After the addition of buffer PL, no clumps should be visible in the sample mixture. Because clumped tissue may not lyse appropriately and therefore leads to a low yield of DNA, homogenization by vortexing or pipetting should be carried out for good result. For typical preparations from leaf tissue, lysis at 65°C for 10~15 minutes would be sufficient. Occasional mixing by shaking or inverting of sample tube accelerates the lysis of cells. Incubation in shaking water bath or equivalents would be the best. Lysis time can be prolonged depending on the tissue type used, but it may be sufficient to incubate for 10~20 minutes in most case.

Filtration after lysis

After tissue lysis, the lysate has some debris and salt precipitates, and these should be removed from the lysate to avoid clogging of GeneAll® SV column at binding step. In traditional methods, cell debris and salt precipitates are discarded through pelleting by centrifugation. Traditional methods require rapid and accurate handling of samples to prevent the pellets from loosening, and make it so difficult to prepare many samples simultaneously. Moreover in case of some plant samples, the pellets are not formed tightly, and this may lead the DNA preparation to poor result. EzSep™ filter included in GeneAll® Exgene™ Plant SV kit makes the preparation of cleared lysate very simple and easy, and facilitates the simultaneous preparation from multiple samples.

In case of some plants, lysate becomes very viscous or sticky after cell lysis, and this leads to shearing of DNA or clogging of EzSep™ filter. We recommend the optional centrifugation in step 4 in Exgene™ Plant SV mini (page 14) to avoid it.



Elution

Purified DNA can be eluted in low salt buffer or deionized water depending on the downstream applications. Buffer AE contains 0.5 mM EDTA and 10 mM TrisCl, pH 9.0. The volume of elution buffer can be adjusted, but it has to be over the minimum requirement. To get higher concentration of DNA, decrease the volume of elution buffer to minimum. For higher overall yield, increase the volume of elution buffer and repeat the elution step again. Optimal yields may be obtained by eluting twice. The concentration and yield in relation to the volume of eluent is shown below.

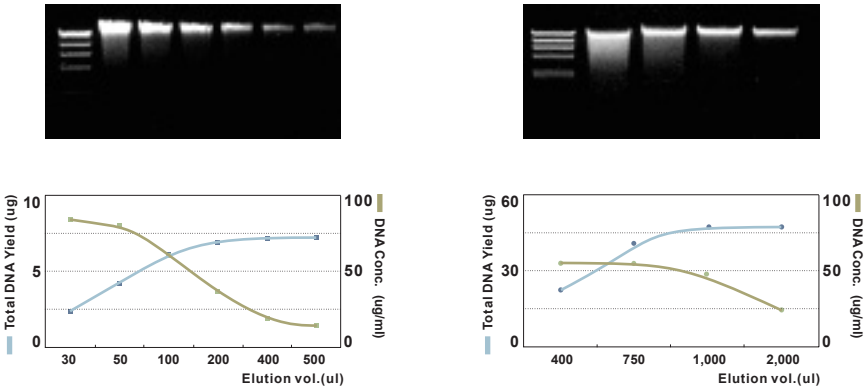


Fig 3. A series of elution volume was applied to DNA purification from 100 mg of pine leaves for mini procedures and 1g of cayenne leaves for MAXI procedures. Each 5 ul of eluate was resolved on 0.8% agarose gel. If the elution volume is reduced for higher concentration of eluate, overall yield will be decreased, especially when the elution volume is below 50 ul for mini, 200 ul for Midi, and 500 ul for MAXI.

Centrifuge in Midi/MAXI kits

GeneAll® Exgene™ Plant SV Midi and MAXI procedures require the conventional centrifuge which has a swinging-bucket rotor and ability of 4,000~5,000 x g. Use of fixed-angle rotor will cause inconsistent contact of SV column membrane with mixtures and/or buffers. Low g-force may lead to incomplete removal of ethanol from SV column membrane. Available centrifuges and rotors are listed below, but you can employ any equivalent.

Company	Centrifuge	Rotor
Beckman Coulter Inc. (California, USA)	Allegra X-15R Allegra 25R	Sx4750 Sx4750A TS-5.1-500
Eppendorf AG (Hamburg, Germany)	5804 / 5804R 5810 / 5810R	A-4-44
EYELA Inc. (Tokyo, Japan)	5800 5900	RS-410 RS-410M
Hanil Science Industrial Inc. (Incheon, Korea)	Union 5KR Union 55R MF-550 HA1000-6 HA1000-3	R-WS1000-6B W-WS750-6B HSR-4S WHSR-4S
Hettich AG (Kirchlengern, Germany)	Rotina 35 Rotanta 460 Rotixa 50S	1717 1724 5624

GeneAll® Exgene™ Plant SV mini

Before experiment

- *Before using for the first time, add absolute ethanol (ACS grade or better) into buffer BD and CW as indicated on the bottle.*
- *Unless there is an other indication, all centrifugation steps should be performed at full speed (>10,000 x g or 10,000~14,000 rpm) in a microcentrifuge at room temperature.*
- *Buffer PL may precipitate upon storage at cold ambient temperature. If so, dissolve it in 65°C water bath.*

■ *Prepare the below;*

» *65°C water bath or heating block*

» *1.5 ml and 2.0 ml microcentrifuge tubes*

» *Microcentrifuge*

1. Grind fresh or frozen plant tissue to a fine powder quickly and completely, using a mortar and pestle pre-cooled with liquid nitrogen. Place up to 100 mg (wet) or 25 mg (dried) of ground tissue into a 1.5 ml or 2.0 ml microcentrifuge tube.

Quick and complete disruption of tissue is essential for good result in preparation. Grinding under liquid nitrogen is the best method for good result, however other methods such as bead-beater or rotor-stator homogenizer can be a good alternative. Lyophilized tissue can be ground at room temperature.

2. Add 400 ul of buffer PL and 4 ul of RNase A solution (100 mg/mL, provided). Vortex vigorously.

Any clumps should not be visible. Mix the lysate by pipetting or vortexing to remove any tissue clumps.

3. Incubate for 10~15 min at 65°C. Mix 2~3 times during incu-bation by inverting or vortexing.

Occasional mixing will accelerate the lysis.

4. Add 140 ul of buffer PD to the lysate. Vortex to mix, and incubate for 5 min on ice.

***(Optional :)* Centrifuge for 5 min at full speed (>10,000 x g or 14,000 rpm).**

For some plants, the lysate becomes very viscous or sticky after addition of buffer PD, and this leads to shearing of DNA or clogging of EzSep™ filter. In this case, removal of precipitates by optional centrifugation will be helpful before proceeding to next step.

5. Apply the lysate to the EzSep™ filter (blue) and centrifuge for 2 min at full speed.

It may be requisite to use [Wide-bore Tip] or to cut the end off the pipet tip to apply the viscous lysate to the EzSep™ filter. Small pellet can be formed in the collection tube after centrifugation. Be careful not to disturb this pellet in next step 6.

6. Transfer the pass-through to a new 1.5 ml microcentrifuge tube by pipetting or decanting carefully not to disturb the cell debris pellet.

About 450 ul of lysate is recovered typically. Recovered volume of lysate can be varied depending on the plant tissue used. Check the correct volume of lysate for optimal binding condition in next step.

7. Add 1.5 volumes of buffer BD to the lysate and mix immediately by pipetting or inverting.

Adjust the volume of buffer BD on the basis of correct volume of lysate. For 450 ul lysate, add 675 ul buffer BD. Immediate mixing is important for optimal binding conditions.

A precipitate can be formed after addition of buffer BD but this will not affect the preparation.

8. Apply 700 ul of the mixture from step 7 to the GeneAll® SV column (green) sitting in collection tube. Centrifuge for 30 sec, and discard the pass-through. Reuse the collection tube.

Any precipitate which may have formed in mixture should be included in transfer.

9. Repeat step 8 with remaining sample.

10. Apply 700 ul buffer CW to the SV column, centrifuge for 30 sec and discard the pass-through, and re-insert the SV column to the collection tube.

11. Add 300 ul of buffer CW to the SV column. Centrifuge for 2 min. Transfer carefully the SV column to a new 1.5 ml microcentrifuge tube (not provided).

Care must be taken at the removal of GeneAll® SV column from the collection tube so the column does not come into contact with the pass-through fraction, as this will result in carryover of ethanol.

Residual ethanol in eluate may interfere with the subsequent reactions. If carryover of buffer CW occurs, centrifuge again for 1 min before proceeding to next step.

12. Add 100 ul of buffer AE directly onto the center of SV column membrane. Incubate for 5 min at room temperature and centrifuge for 1 min.

Elution volume can be decreased to 50 ul for high concentration of DNA, but this will slightly decrease in overall DNA yield. If maximum recovery of DNA is preferred or the starting materials contain large amount of DNA, elution can be done in 200 ul of buffer AE.

13. Repeat step 12.

More 20~40% DNA can be obtained by repeat of eluting.

A new 1.5 ml microcentrifuge tube can be used to prevent dilution of the first eluate.

GeneAll® Exgene™ Plant SV Midi

Before experiment

- *Before using for the first time, add absolute ethanol (ACS grade or better) into buffer BD and CW as indicated on the bottle.*
- *All centrifugation should be performed at room temperature.*
- *Buffer PL may precipitate upon storage at cold ambient temperature. If so, dissolve it in 65°C water bath.*

■ *Prepare the below;*

- *» 65°C water bath or heating block*
- *» 15 ml conical tubes*
- *» Centrifuge capable of 4,000~5,000 x g, which has a swinging-bucket rotor (See page 12)*
- *» The equipment and reagent for tissue disruption; Liquid nitrogen, mortar and pestle*

1. Grind fresh or frozen plant tissue to a fine powder quickly and completely, using a mortar and pestle pre-cooled with liquid nitrogen. Place up to 400 mg (wet) or 100 mg (dried) of ground tissue into a 15 ml conical tube.

Quick and complete disruption of tissue is essential for good result in preparation. Grinding under liquid nitrogen is the best method for good result, however other methods such as bead-beater or rotor-stator homogenizer can be good alternatives. Lyophilized tissue can be ground at room temperature.

2. Add 2 ml of buffer PL and 15 ul of RNase A solution (100 mg/mL, provided). Vortex vigorously.

Any clumps should not be visible. Mix the lysate by pipetting or vortexing to remove any tissue clumps.

3. Incubate for 15~20 min at 65°C. Mix 3~4 times during incu-bation.

Occasional mixing will accelerate the lysis.

4. **Add 700 ul of buffer PD to the lysate. Vortex to mix, and incubate for 10 min on ice.**

5. **Centrifuge for 5 min at 4,000 x g and carefully decant or pipet the supernatant to the EzSep™ Midi filter (green ring).**
   Some debris or salt precipitates can be co-transferred.

6. **Centrifuge for 5 min at 4,000 x g. Transfer the filtrate to a new 15 ml conical tube by pipetting or decanting carefully not to disturb the cell debris pellet.**
   Typically about 2.5 ml of lysate is recovered. Recovered volume of lysate can be varied depending on the plant tissue used. Check the correct volume of lysate for optimal binding condition in next step.

7. **Add 1.5 volumes of buffer BD to the lysate and mix by pipetting or inverting.**
   Adjust the volume of buffer BD on the basis of correct volume of recovered lysate. For 2.5 ml lysate add 3.75 ml buffer BD. Immediate mixing is important for optimal binding conditions.
   A precipitate can be formed after addition of buffer BD but this will not affect the preparation.

8. **Apply 4 ml of the mixture including any precipitate which may have formed from step 7 to GeneAll® SV Midi column (white ring). Centrifuge for 2 min at 4,000 x g, discard the filtrate, and reinsert the SV Midi column to the 15 ml conical tube.**
   Any precipitate which may have formed in mixture should be included in transfer.

9. **Repeat step 8 with the remaining sample.**

10. **Apply 4.5 ml of buffer CW to the SV Midi column, centrifuge for 2 min at 4,000 x g and discard the filtrate, and re-insert the SV Midi column to the 15 ml conical tube.**

11. Add 2 ml buffer CW to the SV Midi column. Centrifuge for 15 min at 4,500 x g. Transfer the SV Midi column to a new 15 ml conical tube (not provided).

Care must be taken at the removal of GeneAll® SV Midi column from the collection tube so the SV column does not come into contact with the pass-through fraction, as this will result in carryover of ethanol.

Residual ethanol in eluate may interfere with the subsequent reactions. If carryover of ethanol occurs, incubate the Midi column for 15 min at RT to evaporate residual ethanol.

12. Add 300 ul of buffer AE directly onto the center of SV Midi column membrane. Incubate for 5 min at room temperature and centrifuge for 5 min at 4,000~5,000 x g.

Elution volume can be decreased to 200 ul for high concentration of DNA, but this will slightly decrease in overall DNA yield.

13. A. For higher concentration of eluate; re-load the eluate from step 12 into the SV Midi column, incubate 5 min at room temperature, and centrifuge for 5 min at 4,000~5,000 x g.

B. For higher overall yield; add 300 ul of fresh buffer AE into the SV Midi column, incubate 5 min at room temperature, and centrifuge for 5 min at 4,000~5,000 x g.

The first and second eluate can be combined or collected separately as necessity.

Less than 300 ul of eluate will be obtained from 300 ul of elution buffer, but this has no influence on DNA yields.

GeneAll® Exgene™ Plant SV MAXI

Before experiment

- *Before using for the first time, add absolute ethanol (ACS grade or better) into buffer BD and CW as indicated on the bottle.*
- *All centrifugation should be performed at room temperature.*
- *Buffer PL may precipitate upon storage at cold ambient temperature. If so, dissolve it in 65°C water bath.*

■ *Prepare the below;*

» *65°C water bath or heating block*

» *50 ml conical tubes*

» *Centrifuge capable of 4,000~5,000 x g, which has a swinging-bucket rotor (See page 12)*

» *The equipment and reagent for tissue disruption; Liquid nitrogen, mortar and pestle*

1. **Grind fresh or frozen plant tissue to a fine powder quickly and completely, using a mortar and pestle pre-cooled with liquid nitrogen. Place up to 1000 mg (wet) or 250 mg (dried) of ground tissue into a 50 ml conical tube.**
   Quick and complete disruption of tissue is essential for good result in preparation. Grinding under liquid nitrogen is the best method for good result, however other methods such as bead-beater or rotor-stator homogenizer can be good alternatives. Lyophilized tissue can be ground at room temperature.

2. **Add 5 ml of buffer PL and 40 ul of RNase A solution (100 mg/mL, provided). Vortex vigorously.**
   No clumps should be visible. Mix the lysate by pipetting or vortexing to remove any tissue clumps.

3. **Incubate for 20 min at 65°C. Mix 3~4 times during incubation time.**

4. **Add 1.8 ml of buffer PD to the lysate. Vortex to mix, and incubate for 10 min on ice.**

5. **Centrifuge for 5 min at 4,000 x g and carefully decant or pipet the supernatant to the EzSep™ MAXI filter (blue).**
   Some debris or salt precipitates can be co-transferred.

6. **Centrifuge for 5 min at 4,000 x g and transfer the pass-through to a new 50 ml conical tube by pipetting or decanting carefully not to disturb the cell debris pellet.**
   Typically, 5~6 ml of lysate is recovered. Recovered volume of lysate can be varied depending on the plant tissue used. Check the correct volume of lysate for optimal binding condition in next step.

7. **Add 1.5 volumes of buffer BD to the lysate and mix by pipetting or inverting.**
   Adjust the volume of buffer BD on the basis of correct volume of recovered lysate. For 5 ml lysate add 7.5 ml buffer BD. Immediate mixing is important for optimal binding conditions.
   A precipitate can be formed after addition of buffer BD but this will not affect the preparation.

8. **Apply the sample mixture including any precipitate which may have formed from step 7 to the GeneAll® SV MAXI column (white). Centrifuge for 2 min at 4,000 x g and discard the pass-through and re-insert the MAXI column to the collection tube.**

9. **Apply 13 ml of buffer CW to the SV MAXI column, centrifuge for 2 min at 4,000 x g and discard the pass-through, and re-insert the SV MAXI column to the collection tube.**

10. Add 5 ml buffer CW to the SV MAXI column. Centrifuge for 15 min at 4,500 x g. Transfer the SV MAXI column to a new 50 ml conical tube (not provided).

Care must be taken at the removal of GeneAll® SV MAXI column from the collection tube so the MAXI column does not come into contact with the pass-through fraction, as this will result in carryover of ethanol. Residual ethanol in eluate may interfere with the subsequent reactions. If carryover of ethanol occurs, incubate the MAXI column for 15 min at RT to evaporate residual ethanol.

11. Add 0.6~1 ml of buffer AE directly onto the center of SV MAXI column membrane. Incubate for 5 min at room temperature and centrifuge for 5 min at 4,000~5,000 x g.

Elution volume can be decreased to 500 ul for high concentration of DNA, but this will slightly decrease in overall DNA yield.

12. A. For higher concentration of eluate; re-load the eluate from step 11 into the SV MAXI column, incubate 5 min at room temperature, and centrifuge for 5 min at 4,000~5,000 x g.

B. For higher overall yield; add 0.6~1 ml of fresh buffer AE into the SV MAXI column, incubate 5 min at room temperature, and centrifuge for 5 min at 4,000~5,000 x g.

The first and second eluates can be combined or collected separately as necessity.

Troubleshooting Guide

Facts	Possible Causes	Suggestions
Low or no recovery	**Too much starting material**	Too much starting material lead to inefficient lysis and column clogging, followed by poor DNA yields. Reduce the amount of starting material.
	Too old or mis-stored sample used	Refer "Sample preparation, pulverization and lysis"on page 9.
	Insufficient pulverization	Refer "Sample preparation, pulverization and lysis" on page 9.
	Incorrect binding	Ensure the binding conditions are adjusted correctly in step 7.
	Improper elution	The condition for optimal elution is of low salt concentration with weakly alkaline pH (7.0<pH<9.0). Ensure the condition when water or other buffer was used as eluent. After eluent is applied on the center of column membrane, it is essential to incubate at least for 5 minutes at room temperature.
	Improper centrifuge (Midi/MAXI)	Swinging-bucket rotor (capable of 4,000~5,000 x g) should be used fixed-angle rotor is not compatible with this kit (See page 12).
	Incomplete precipitation	Any cell debris or precipitates should be removed before addition of buffer BD.
Low purity	**Insufficient lysis**	Too much starting material can lead to poor lysis, followed by low purity of DNA.
	Improper centrifuge (Midi/MAXI)	Swinging-bucket rotor (capable of 4,000~5,000 x g) should be used fixed-angle rotor is not compatible with this kit (See page 12).

Troubleshooting Guide

Facts	Possible Causes	Suggestions
Clogging of EzSep™ filter	**High viscosity of lysate (mini)**	Perform the optional centrifugation step in step 4 before applying to EzSep™ filter.
	Insufficient centrifugation (Midi/MAXI)	Increase the g-force and centrifugation time (See page 12).
Clogging of GeneAll® Exgene™ Plant SV column	**Incomplete removal of precipitate**	Any cell debris or precipitates should be removed before addition of buffer BD.
	Lysate too viscous or sticky	Reduce the amount of starting sample, or increase the amount of buffer PL and PD.
	Insufficient centrifugation (Midi/MAXI)	Increase the g-force and centrifugation time (See page 12).
DNA sheared	**Too much starting materials**	Too much starting material can make the lysate very viscous and lead to shearing of DNA. Reduce the amount of starting material.
	Too old or mis-stored sample used	Refer "Sample preparation, pulverization and lysis"on page 9.
	Too viscous lysate (mini)	In some plants, the lysate may become too viscous, so the optional centrifugation in step 4 should be performed before applying to EzSep™ filter.
Enzymatic reaction is not performed well with purified DNA	**High salt concentration in eluate**	Ensure that washing step was carried out just in accordance with the protocols. Repeat of washing step may help to remove high salt in eluate.
	Low purity of DNA	See "Low purity" at page 22.
	Residual ethanol in eluate	Ensure that the wash step in protocols is performed properly. GeneAll® Exgene™ Plant SV column membrane should be completely dried by additional centrifugation or air-drying before elution.

Ordering Information

GeneAll® *Hybrid-Q™* *for rapid preparation of plasmid DNA*

Products	Scale	Size	Cat. No.	Type
Plasmid Rapidprep	mini	50	100-150	spin
		200	100-102	

GeneAll® *Exprep™* *for preparation of plasmid DNA*

Products	Scale	Size	Cat. No.	Type
Plasmid SV	mini	50	101-150	spin / vacuum
		200	101-102	
	Midi	26	101-226	spin / vacuum
		50	101-250	
		100	101-201	

GeneAll® *Exfection™* *for preparation of transfection-grade plasmid DNA*

Products	Scale	Size	Cat. No.	Type
Plasmid LE *(Low Endotoxin)*	mini	50	111-150	spin / vacuum
		200	111-102	
	Midi	26	111-226	spin / vacuum
		100	111-201	
Plasmid EF *(Endotoxin Free)*	Midi	20	121-220	spin
		100	121-201	

GeneAll® *Expin™* *for purification of fragment DNA*

Products	Scale	Size	Cat. No.	Type
Gel SV	mini	50	102-150	spin / vacuum
		200	102-102	
PCR SV	mini	50	103-150	spin / vacuum
		200	103-102	
CleanUp SV	mini	50	113-150	spin / vacuum
		200	113-102	
Combo GP	mini	50	112-150	spin / vacuum
		200	112-102	

GeneAll® *Exgene™* *for isolation of total DNA*

Products	Scale	Size	Cat. No.	Type
Tissue SV	mini	100	104-101	spin / vacuum
		250	104-152	
	Midi	26	104-226	spin / vacuum
		100	104-201	
	MAXI	10	104-310	spin / vacuum
		26	104-326	
Tissue *plus!* SV	mini	100	109-101	spin / vacuum
		250	109-152	
	Midi	26	109-226	spin / vacuum
		100	109-201	
	MAXI	10	109-310	spin / vacuum
		26	109-326	

GeneAll® *Exgene™* *for isolation of total DNA*

Products	Scale	Size	Cat. No.	Type
Blood SV	mini	100	105-101	spin / vacuum
		250	105-152	
	Midi	26	105-226	spin / vacuum
		100	105-201	
	MAXI	10	105-310	spin / vacuum
		26	105-326	
Cell SV	mini	100	106-101	spin / vacuum
		250	106-152	
	MAXI	10	106-310	spin / vacuum
		26	106-326	
Clinic SV	mini	100	108-101	spin / vacuum
		250	108-152	
	Midi	26	108-226	spin / vacuum
		100	108-201	
	MAXI	10	108-310	spin / vacuum
		26	108-326	
Genomic DNA micro		50	118-050	spin
Plant SV	mini	100	117-101	spin / vacuum
		250	117-152	
	Midi	26	117-226	spin / vacuum
		100	117-201	
	MAXI	10	117-310	spin / vacuum
		26	117-326	
Soil DNA mini	mini	50	114-150	spin
Stool DNA mini	mini	50	115-150	spin
Viral DNA / RNA	mini	50	128-150	spin
FFPE Tissue DNA	mini	50	138-150	spin
		250	138-152	

GeneAll® *GenEx™* *for isolation of total DNA without spin column*

Products	Scale	Size	Cat. No.	Type
GenEx™ Blood	Sx	100	220-101	solution
		500	220-105	
	Lx	100	220-301	solution
GenEx™ Cell	Sx	100	221-101	solution
		500	221-105	
	Lx	100	221-301	solution
GenEx™ Tissue	Sx	100	222-101	solution
		500	222-105	
	Lx	100	222-301	solution

GeneAll® *GenEx™* *for isolation of total DNA*

Products	Scale	Size	Cat. No.	Type
GenEx™ Plant	Sx	100	227-101	solution
	Mx	100	227-201	
	Lx	100	227-301	
GenEx™ Plant *plus!*	Sx	100	228-101	solution
	Mx	50	228-250	
	Lx	20	228-320	

GeneAll® *DirEx™* series

for preperation of PCR-template without extraction

Products	Scale	Size	Cat. No.	Type
DirEx™		100	250-101	solution
DirEx™ *Fast*-Tissue		96 T	260-011	solution
DirEx™ *Fast*-Cultured cell		96 T	260-021	solution
DirEx™ *Fast*-Whole blood		96 T	260-031	solution
DirEx™ *Fast*-Blood stain		96 T	260-041	solution
DirEx™ *Fast*-Hair		96 T	260-051	solution
DirEx™ *Fast*-Buccal swab		96 T	260-061	solution
DirEx™ *Fast*-Cigarette		96 T	260-071	solution

GeneAll® RNA series *for preperation of total RNA*

Products	Scale	Size	Cat. No.	Type
RiboEx™	mini	100	301-001	solution
		200	301-002	
Hybrid-R™	mini	100	305-101	spin
Hybrid-R™ Blood RNA	mini	50	315-150	spin
Hybrid-R™ miRNA	mini	50	325-150	spin
RiboEx™ LS	mini	100	302-001	solution
		200	302-002	
Riboclear™	mini	50	303-150	spin
Riboclear™ *plus!*	mini	50	313-150	spin
Ribospin™	mini	50	304-150	spin
Ribospin™ II	mini	50	314-150	spin
		300	314-103	
Ribospin ™ vRD	mini	50	302-150	spin
Ribospin ™ vRD *plus!*	mini	50	312-150	spin
Ribospin ™ vRD II	mini	50	322-150	spin
Ribospin ™ Plant	mini	50	307-150	spin
Ribospin™ Seed / Fruit	mini	50	317-150	spin
Allspin™	mini	50	306-150	spin
RiboSaver™	mini	100	351- 001	solution

GeneAll® *AmpONE™* *for PCR amplification*

Products	Scale	Size	Cat. No.	Type
Taq DNA polymerase		250 U	501-025	(2.5 U/㎕)
		500 U	501-050	
		1,000 U	501-100	
α-Taq DNA polymerase		250 U	502-025	(2.5 U/㎕)
		500 U	502-050	
		1,000 U	502-100	
α-Pfu DNA polymerase		250 U	504-025	(2.5 U/㎕)
		500 U	504-050	
		1,000 U	504-100	
Fast-Pfu DNA polymerase		250 U	505-025	(2.5 U/㎕)
		500 U	505-050	
		1,000 U	505-100	
Hotstart Taq DNA polymerase		250 U	531-025	(2.5 U/㎕)
		500 U	531-050	
		1,000 U	531-100	
Taq Premix	96 tubes	20 ㎕	521-200	lyophilized
		50 ㎕	521-500	
		20 ㎕	526-200	solution
		50 ㎕	526-500	
α-Taq Premix	96 tubes	20 ㎕	522-200	lyophilized
		50 ㎕	522-500	
		20 ㎕	527-200	solution
		50 ㎕	527-500	
HS-Taq Premix	96 tubes	20 ㎕	525-200	solution
		50 ㎕	525-500	
		20 ㎕	520-200	lyophilized
α-Pfu Premix	96 tubes	50 ㎕	523-500	solution
Taq Premix (w/o dye)	96 tubes	20 ㎕	524-200	lyophilized
dNTPs mix		500 ㎕	509-020	2.5 mM each
dNTPs set (set of dATP, dCTP, dGTP and dTTP)		1 ml x 4 tubes	509-040	100 mM

Products	Scale	Size	Cat. No.	Type

GeneAll® *AmpMaster™* *for PCR amplification*

Taq Master mix	0.5 ml x 2 tubes	541-010	solution
	0.5 ml x 10 tubes	541-050	solution
α-Taq Master mix	0.5 ml x 2 tubes	542-010	solution
	0.5 ml x 10 tubes	542-050	solution
HS-Taq Master mix	0.5 ml x 2 tubes	545-010	solution
	0.5 ml x 10 tubes	545-050	solution
α-Pfu Master mix	0.5 ml x 2 tubes	543-010	solution
	0.5 ml x 10 tubes	543-050	solution

GeneAll® *HyperScript™* *for Reverse Transcription*

Reverse Transcriptase	10,000 U	601-100	solution
RT Master mix	0.5 ml × 2 tubes	601-710	solution
RT Master mix with oligo $(dT)_{20}$	0.5 ml × 2 tubes	601-730	solution
RT Master mix with random hexamer	0.5 ml × 2 tubes	601-740	solution
RT Premix	96 tubes, 20 $\mu\ell$	601-602	solution
RT Premix with oligo $(dT)_{20}$	96 tubes, 20 $\mu\ell$	601-632	solution
RT Premix with random hexamer	96 tubes, 20 $\mu\ell$	601-642	solution
One-step RT-PCR Master mix	0.5 ml × 2 tubes	602-110	solution
One-step RT-PCR Premix	96 tubes, 20 $\mu\ell$	602-102	solution
First strand Synthesis Kit	50 reaction	605-005	solution
ZymAll™ RNase Inhibitor	10,000 U	605-010	solution
ZymAll™ RNase Inhibitor	4,000 U	605-004	solution

GeneAll® **RealAmp™** *for qPCR amplification*

SYBR qPCR Master mix (2X, Low ROX)	200 rxn 20 $\mu\ell$	801-020	solution
	500 rxn 20 $\mu\ell$	801-050	
SYBR qPCR Master mix (2X, High ROX)	200 rxn 20 $\mu\ell$	801-021	solution
	500 rxn 20 $\mu\ell$	801-051	

Products	Size	Cat. No.	

GeneAll® **Protein series**

ProtinEx™ Animal cell / tissue	100 ml	701-001	solution
PAGESTA™ Reducing 5X SDS-PAGE Sample Buffer	1 ml × 10 tubes	751-001	solution

GeneAll® STEADi™ *for automatic nucleic acid puritication*

STEADi™	12 Instrument		GST012
STEADi™	24 Instrument		GST024
STEADi™	Genomic DNA Cell / Tissue Kit	96	401-104
STEADi™	Genomic DNA Blood Kit	96	402-105
STEADi™	Bacteria DNA Kit	96	403-106
STEADi™	Total RNA Kit	96	404-304
STEADi™	Viral DNA / RNA Kit	96	405-322
STEADi™	CFC Seed DNA / RNA Kit	96	406-C02

GeneAll

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