2009

PLANT SV MINI
PLANT SV MIDI
PLANT SV MAXI

DNA PURIFICATION HANDBOOK

exgene™

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	100
GeneAll® SV column type G	100	250	26	100
EzSep™ filter column	100	250	26	100
Collection tube	200	500	52	200
Buffer PL	100 ml	200 ml	100 ml	300 ml
Buffer PD	30 ml	90 ml	30 ml	120 ml
Buffer BD	110 ml	310 ml	110 ml	400 ml
Buffer CW	150 ml	400 ml	250 ml	1000 ml
Buffer AE*	60 ml	120 ml	60 ml	120 ml
RNase A (100 mg/ml)	0.48 ml	1.3 ml	0.48 ml	1.8 ml
Protocol Handbook	1	1	1	1

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

	Plant SV MAXI	
Cat. No.	**117-310**	**117-326**
Size	**MAXI**	**MAXI**
No. of preparation	10	26
GeneAll® SV column type G	10	26
EzSep™ filter column	10	26
Collection tube	20	52
Buffer PL	100 ml	200 ml
Buffer PD	30 ml	90 ml
Buffer BD	110 ml	310 ml
Buffer CW	250 ml	750 ml
Buffer AE*	60 ml	120 ml
RNase A (100 mg/ml)	0.48 ml	1.3 ml
Protocol Handbook	1	1

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

GeneAll® Exgene™ Plant SV	mini	Midi*	MAXI*
Sample size	~ 100 mg wet	~ 400 mg wet	~ 1g 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 swining-bucket rotor and ability of 4,000 ~ 5,000 xg.*

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.

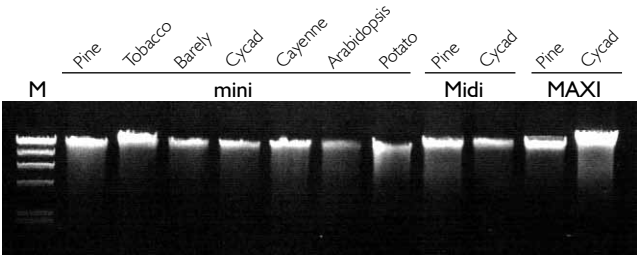


Fig 1. Genomic DNA prepared from various plant tissues using GeneAll® Exgene™ Plant SV series. Purified DNA was resolved on 0.7 % agarose gel.

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.*

Starting sample size

There is an optimized sample size 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 size of sample, GeneAll® Exgene™ Plant SV Midi/MAXI is available.

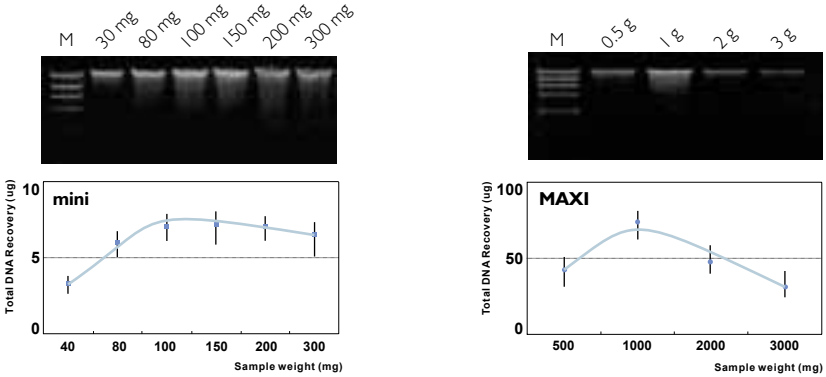


Fig 2. DNA Preparation from pine.
Use of an excessive starting sample may cause incomplete lysis of sample tissues and the shearing of DNA, resulting in low yield and poor quality of DNA. 2 ul out of 100 ul eluate was resolved on 0.8 % agarose gel. M : Lambda-HindIII

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.

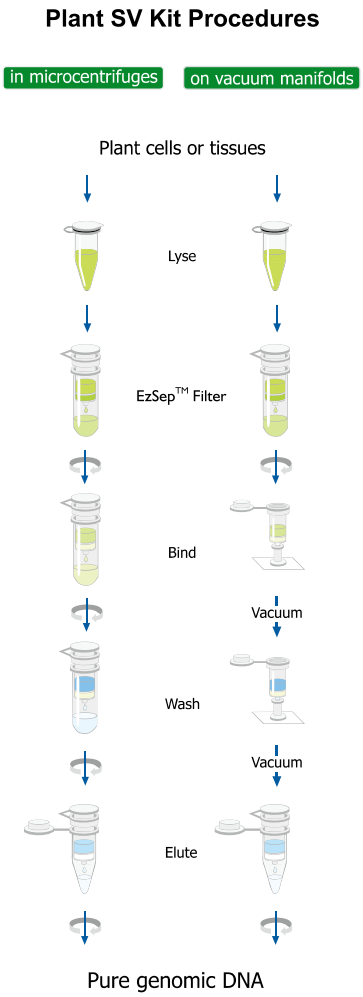
Before lysis, tissue sample should be disrupted completely for efficient lysis, and this step should be performed at low temperature (below 0°C) as quickly as possible for optimized result. Lyophilized tissue can be ground at ambient condition.
Mortar and pestle with liquid nitrogen is a typical and 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 (mini) to avoid it.



Elution

Purified DNA can be eluted in low salt buffer or deionized water depending on the downstream applications. Buffer AE contains 0.5mM EDTA and 10mM 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.

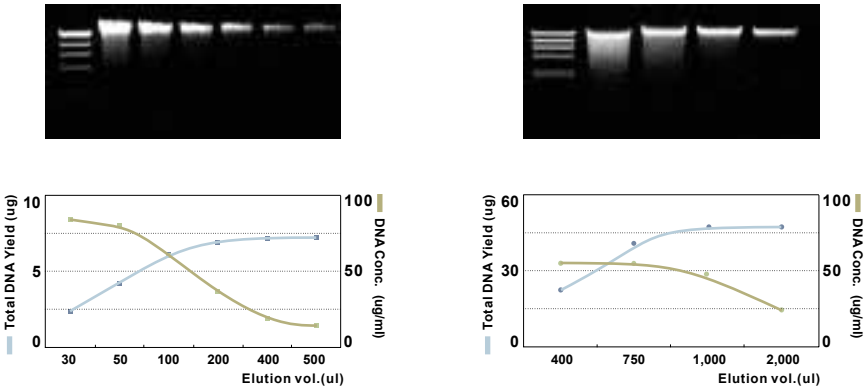


Fig 3. A series of elution volume was applied to DNA purification from 100 mg of pine leaves for mini procedures and 1g of cayenne leaves for MAXI procedures. Each 5 ul of eluate was resolved on 0.8 % agarose gel. If the elution volume is reduced for higher concentration of eluate, overall yield will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

Unless there is an other indication, all centrifugation steps should be performed at full speed (>10,000 xg 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 mL Micro centrifuge 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 ml 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 incubation 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 xg 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 tube by pipetting or decanting carefully not to disturb the cell debris pellet.

About 450 ul of lysate is recovered typically. Recoverd 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 vol 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 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 tube can be used to prevent dilution of the first eluate.

GeneAll® Exgene™ Plant SV Midi

Before experiment

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 xg, 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 incubation.**
   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 xg 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 xg. Transfer the filtrate to a new 15 ml tube by pipetting or decanting carefully not to disturb the cell debris pellet.**
   Typically about 2.5 ml of lysate is recovered. Recoverd 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 vol 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 xg, discard the filtrate, and reinsert the SV Midi Column to the 15 ml 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 xg and discard the filtrate, and re-insert the SV Midi Column to the 15 ml tube.**

11. Add 2 ml Buffer CW to the SV Midi Column. Centrifuge for 15 min at 4,500 xg. Transfer the SV Midi Column to a new 15 ml 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 xg.

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 xg.

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 xg.

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

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 xg, 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 xg 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 xg and transfer the pass-through to a new 50 ml tube by pipetting or decanting carefully not to disturb the cell debris pellet.**
   Typically, 5 ~ 6 ml of lysate is recovered. Recoverd 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 vol 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 xg 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 xg 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 xg. 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 xg.

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 xg.

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 xg.

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, disruption and lysis"on page 9.
	Insufficient pulverization	Refer "Sample preparation, disruption 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<pH<9). 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)	Swing-bucket rotor (capable of 4,000 ~ 5,000 xg) 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)	Swing-bucket rotor (capable of 4,000 ~ 5,000 xg) should be used fixed-angle rotor is not compatible with this kit (See page 12).

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, disruption 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 23.
	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

Products	Size	Type	Cat. No.
GeneAll® *Exprep™* *for preparation of plasmid DNA*			
Plasmid SV mini	50 200 1,000	spin / vacuum	101-150 101-102 101-111
Plasmid SV Midi**	26 50 100	spin / vacuum	101-226 101-250 101-201
Plasmid SV Quick	50 200 1,000	mini / spin	101-050 101-002 101-011
GeneAll® *Exfection™* *for preparation of highly pure plasmid DNA*			
Plasmid LE mini *(Low Endotoxin)*	50 200	spin / vacuum	111-150 111-102
Plasmid LE Midi* *(Low Endotoxin)*	26 100	spin / vacuum	111-226 111-201
Plasmid EF Midi* *(Endotoxin Free)*	20 100	spin	121-220 121-201
GeneAll® *Expin™* *for purification of fragment DNA*			
Gel SV	50 200	mini / spin / vacuum	102-150 102-102
PCR SV	50 200	mini / spin / vacuum	103-150 103-102
CleanUp SV	50 200	mini / spin / vacuum	113-150 113-102
Combo GP	50 200	mini / spin / vacuum	112-150 112-102
GeneAll® *Exgene™* *for isolation of total DNA*			
Tissue SV mini (*plus!*)*	100 250	spin / vacuum	104(9)-101 104(9)-152
Tissue SV Midi (*plus!*)**	26 100	spin / vacuum	104(9)-226 104(9)-201
Tissue SV MAXI (*plus!*)**	10 26	spin / vacuum	104(9)-310 104(9)-326
Blood SV mini	100 250	spin / vacuum	105-101 105-152
Blood SV Midi**	26 100	spin / vacuum	105-226 105-201
Blood SV MAXI**	10 26	spin / vacuum	105-310 105-326
Cell SV mini	100 250	spin / vacuum	106-101 106-152
Cell SV MAXI**	10 26	spin / vacuum	106-310 106-326
Clinic SV mini	100 250	spin / vacuum	108-101 108-152

Products	Size	Type	Cat. No.
GeneAll® *Exgene™* *for isolation of total DNA*			
Clinic SV Midi	26 100	spin / vacuum	108-226 108-201
Clinic SV MAXI**	10 26	spin / vacuum	108-310 108-326
Plant SV mini	100 250	spin / vacuum	117-101 117-152
Plant SV Midi**	26 100	spin / vacuum	117-226 117-201
Plant SV MAXI**	10 26	spin / vacuum	117-310 117-326
GMO SV mini	50 200	spin / vacuum	107-150 107-102
GeneAll® *GenEx™* *for isolation of total DNA*			
Genomic Sx† Genomic Sx† Genomic Lx††	100 500 100	mini / solution mini / solution MAXI / solution	208-001 208-005 208-301
GeneAll® *RiboEx™* *for preparation of total RNA*			
RiboEx™	100 200	solution	301-001 301-002
RiboEx™_column	50	spin	301-150
RiboEx™ LS	100 200	solution	302-001 302-002
Ribo_clear™	50	spin	303-150
GeneAll® *AmpONE™* *for PCR amplification*			
Taq DNA polymerase	250 U 500 U 1,000 U	(2.5 U/㎕)	501-025 501-050 501-100
α-Taq DNA polymerase	250 U 500 U 1,000 U	(2.5 U/㎕)	502-025 502-050 502-100
Pfu DNA polymerase	250 U 500 U 1,000 U	(2.5 U/㎕)	503-025 503-050 503-100
Taq Master mix	2x	0.5 ml x 2 tubes	511-010
α-Taq Master mix	2x	0.5 ml x 2 tubes	512-010
Taq Premix	20 ㎕ 50 ㎕	96 tubes	521-200 521-500
α-Taq Premix	20 ㎕ 50 ㎕	96 tubes	522-200 522-500
dNTP mix	500 ㎕	2.5 mM each	509-020
dNTP set (set of dATP, dCTP, dGTP and dTTP)	1 ml x 4 tubes	100 mM	509-040

* Each dNTP is available

** GeneAll® Tissue SV mini, Midi, and MAXI plus! kit provide the additional methods for the purification from animal whole blood.*

*** GeneAll® SV Midi / MAXI kits require the centrifuge which has a swinging-bucket rotor and ability of 4,000 ~ 5,000 xg.*

† On the basis of DNA purification from 300 ul whole blood.

†† On the basis of DNA purification from 10 ml whole blood.

GeneAll

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