Ver. 1903-00

HelixAmp™ Direct WGA Kit

Kit Contents

HelixAmp™ Direct WGA Kit		
Cat. No.	**DWGA50** (50 rxns/kit)	**DWGA100** (100 rxns/kit)
1 M DTT	250 µl	500 µl
PBS Buffer	50 µl	100 µl
DB [WGAD]	1.25 ml x 2ea	1.25 ml x 4ea
NB [WGAD]	100 µl	200 µl
Primer Mix [WGA]	50 µl	100 µl
Enzyme Mix [WGA]	50 µl	100 µl
Reaction Buffer [WGA]	0.6 ml	1.2 ml
dNTP Mix (each 10 mM)	100 µl	200 µl
Instruction for Use	1ea	1ea

*** Store at -20℃**

Description

HelixAmp™ Direct WGA Kit is a complete system for whole genome amplification from various tissues or samples directly without DNA purification processes. Very little amount of samples, several milligram or microliter volume, are required for the direct WGA. About 10 µg DNA products could be obtained in a standard reaction. The enzyme mix and buffer system are designed to tolerate against most amplification inhibitors found in crude samples. *Phi29* DNA polymerase, the major polymerization enzyme of this kit, isothermally amplifies the genomic DNAs included in the samples with multiple displacement mechanism. *Phi29* DNA polymerase could produce DNA strand up to 70 kb long with high fidelity. All required components including enzymes, buffers, dNTPs, random primers, and sample pretreatment reagents are supplied in this kit. The amplified DNA products could be applied for successive PCR, genotyping, and library construction.

- Fast and uniform amplification across entire genome
- Multiple Displacement Amplification by *Phi29* DNA polymerase
- Direct WGA from Whole blood, animal tissues, plant leaves and seeds, and clinical & forensic sample [Saliva, Buccal swab, Hair root, Blood stain(toilet paper or paper)]

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Application

Genotype analysis
PCR and real-time PCR
Construction of genomic library

Quality control assay data

Functional assay

HelixAmp™ Direct WGA Kit was tested for the direct amplification from 1 μl of blood sample.

Evaluation of amplified DNA

- **Agarose gel electrophoresis** : The integrity of the amplified DNA is assessed by agarose gel electrophoresis.
- **Quantification of amplified DNA** : The DNA amplified with HelixAmp™ Genome Amplifier are quantified using a quantitative Real-time PCR Assay.

Quality authorized by Yountaek Go 

Protocol

<Blood>

1. **Prepare the DM Buffer by mixing as follows.**

DM Buffer	
DB [WGAD]	5.0 μl * N
1 M DTT	0.5 μl * N

N : number of reactions

※ Caution : DM buffer should be freshly prepared before use.

2. **Sample preparation.**

   1) Add **1 μl** of **PBS Buffer** to **0.5~1 μl** of **Whole Blood** sample.
   2) Add **1.5 μl** of **DM Buffer** and mix by pipetting.
   3) Incubate at ice for 10 min.
   4) Add **1.5 μl** of **NB [WGAD]**. Briefly vortex and spin down.

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3. **Add followings and mix well to make final 20 µl reaction mixture.**

Reaction buffer [WGA]	12 µl
dNTP Mix (each 10mM)	2 µl
Primer Mix [WGA]	1 µl
Enzyme Mix [WGA]	1 µl

4. **Incubate at 30℃ for 1.5 hours and inactivate the enzyme at 65℃ for 3 min.**

   ※ **Perform the reaction at a thermal cycler or incubator. Water-bath is not recommendable. For PCR, use 1 ~ 2 µl of 10-fold diluted product with distilled water. If the PCR is not successful, it is recommended to use 1 ~ 2 µl of undiluted product as PCR template.**

5. **Store amplified DNA at -20℃.**

<Animal tissue>

1. **Prepare the DM Buffer by mixing as follows.**

DM Buffer	
DB [WGAD]	50 µl * N
1 M DTT	5 µl * N

N : number of reactions

※ Caution : DM buffer should be freshly prepared before use.

2. **Sample preparation.**

   1) Transfer **50 µl** of **DM Buffer** into a 1.5 ml microtube.
   2) Add a tissue slice size of about 5 mm into the DM buffer. Briefly mix by vortexing and spin down.
   3) Incubate at Room temperature for 10 min.
   4) Transfer **2 µl** of the **supernatant** into a new 1.5 ml microtube.
   5) Add **2 µl** of **NB [WGAD]**. Mix by pipetting and spin down.

3. **Add followings and mix well to make final 20 µl reaction mixture.**

Reaction buffer [WGA]	12 µl
dNTP Mix (each 10mM)	2 µl
Primer Mix [WGA]	1 µl
Enzyme Mix [WGA]	1 µl

4. **Incubate at 30℃ for 1.5 hours and inactivate the enzyme at 65℃ for 3 min.**
   ※ **Perform the reaction at a thermal cycler or incubator. Water-bath is not recommendable. For PCR, use 1 ~ 2 µl of 10-fold diluted product with distilled water. If the PCR is not successful, it is recommended to use 1 ~ 2 µl of undiluted product as PCR template.**

5. **Store amplified DNA at -20℃.**

<Plant leaves>

1. **Prepare the DM Buffer by mixing as follows.**

DM Buffer	
DB [WGAD]	50 µl * N
1 M DTT	5 µl * N

N : number of reactions

※ Caution : DM buffer should be freshly prepared before use.

2. **Sample preparation.**
   1) Transfer **50 µl** of **DM Buffer** into a 1.5 ml microtube.
   2) Add a plant leaf cut size of about 5 mm into the DM buffer. Briefly mix by pipetting or vortexing, and spin down.
   3) Incubate at room temperature for 10 min.
   4) Transfer **2 µl** of the **supernatant** into a new 1.5 ml microtube.
   5) Add **2 µl** of **NB [WGAD]**. Mix by pipetting and spin down.

3. **Add followings and mix well to make final 20 µl reaction mixture.**

Reaction buffer [WGA]	12 µl
dNTP Mix (each 10mM)	2 µl
Primer Mix [WGA]	1 µl
Enzyme Mix [WGA]	1 µl

4. **Incubate at 30℃ for 1.5 hours and inactivate the enzyme at 65℃ for 3 min.**
   ※ **Perform the reaction at a thermal cycler or incubator. Water-bath is not recommendable. For PCR, use 1 ~ 2 µl of 10-fold diluted product with distilled water. If the PCR is not successful, it is recommended to use 1 ~ 2 µl of undiluted product as PCR template.**

5. **Store amplified DNA at -20℃.**

<Seeds>

1. **Prepare the DM Buffer by mixing as follows.**

DM Buffer	
DB [WGAD]	50 µl * N
1 M DTT	5 µl * N

N : number of reactions

※ Caution : DM buffer should be freshly prepared before use.

2. **Preparation of reaction mixture.**

   1) **50 µl** of **DM Buffer** into a 1.5 ml microtube.
   2) Add several small (<1mm size) pieces of **cracked plant seeds** to the DM Buffer. Mix by vortexing and spin down.
   3) Incubate at Room temperature for 10 min.
   4) Transfer **2 µl** of **supernatant** into a new 1.5 ml microtube.
   5) Add **2 µl** of **NB [WGAD]**. Mix by pipetting and spin down.

3. **Add followings and mix well to make final 20 µl reaction mixture.**

Reaction buffer [WGA]	12 µl
dNTP Mix (each 10mM)	2 µl
Primer Mix [WGA]	1 µl
Enzyme Mix [WGA]	1 µl

4. **Incubate at 30℃ for 1.5 hours and inactivate the enzyme at 65℃ for 3 min.**

   ※ Perform the reaction at a thermal cycler or incubator. Water-bath is not recommendable. For PCR, use 1 ~ 2 µl of 10-fold diluted product with distilled water. If the PCR is not successful, it is recommended to use 1 ~ 2 µl of undiluted product as PCR template.

5. **Store amplified DNA at -20℃.**

Products

Cat. No.	Products	Size
DWGA50	HelixAmp™ Direct WGA Kit	50 rxns
DWGA100	HelixAmp™ Direct WGA Kit	100 rxns

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