GE Healthcare

illustra GenomiPhi V2 DNA Amplification Kit

Product Web Protocol

Codes: 25-6600-30
25-6600-31
25-6600-32

DOMINIQUE DUTSCHER SAS

Contents

1. Legal 3
1.1. Product use restriction/warranty 3

2. Handling, Safety Warnings and Precautions 4
2.1. Handling, preparation, and storage of starting materials 4
2.2. Storage Conditions 4
2.3. Expiry 4

3. Components 5
3.1. Kit Contents 5
3.2. Reagents to be supplied by the user 5

4. Product Description 6
4.1. The Basic Principle 6
4.2. Kit Specifications 6

5. Protocols 8
5.1. Short Protocol 8
5.2. General Protocol 8
5.3. Chemical Denaturation and Amplification of Template DNA 9
5.4. DNA Amplifications from Blood Cells 10
5.5. DNA Amplification from Cheek Cells (Mouth Wash or Buccal Swab) 11
5.6. DNA Amplification from Blood or Buccal Swab-blotted Paper (E.g. Whatman's FTA Paper or Guthrie Card) 13
5.7. Quantitation of Amplification Products 14

6. Appendix 17
6.1. Troubleshooting 17
6.2. References 18
6.3. Related Products 18

DOMINIQUE DUTSCHER SAS

1. Legal

GE and GE Monogram are trademarks of General Electric Company.

Amersham, Autoseq, CyScribe, DYEnamic, GenomiPhi, GFX, MegaBACE, MicroSpin, ProbeQuant, PureTaq and Ready-To-Go are trademarks of GE Healthcare companies.

Nucleon is a trademark of Tepnel Life Sciences Plc. PicoGreen is a trademark of Invitrogen, Inc.

For use only as licensed by Qiagen GmbH and GE Healthcare Bio-sciences Corp. The Phi 29 DNA polymerase enzyme may not be re-sold or used except in conjunction with the other components of the **Genomiphi V2 DNA Amphification Kit.** See U.S. patent Nos. 5,854,033, 6,124,120, 6,143,495, 5,001,050, 5,198,543, 6,323,009, 5,576,204 and related U.S. and foreign patents.

GE Healthcare reserves the right, subject to any regulatory approval, to make changes in specifications and features shown herein, or discontinue the product described at any time without notice or obligation.

Contact your GE Representative for the most current information and a copy of the terms and conditions.

© 2006 General Electric Company - All rights reserved.

http://www.gehealthcare.com/lifesciences
GE Healthcare UK Limited Amersham Place, Little Chalfont, Buckinghamshire, HP7 9NA, UK.

1.1. Product use restriction/warranty

The **GenomiPhi V2 DNA Amplification Kit** components have been designed, developed, and sold **for research purposes only.** They are suitable **for *in vitro* use only.** Not recommended or intended for diagnosis of disease in humans or animals. No claim or representation is intended for its use to identify any specific organism or for clinical use (diagnostic, prognostic, therapeutic, or blood banking). Do not use internally or externally in humans or animals.

It is the responsibility of the user to verify the use of the **GenomiPhi V2 DNA Amplification Kit** for a specific application range as the performance characteristic of this kit has not been verified to a specific organism.

DOMINIQUE DUTSCHER SAS

2. Handling. Safety Warnings and Precautions

2.1. Handling, preparation, and storage of starting materials

This kit is sensitive to small amounts of DNA. Wear gloves and safety glasses to avoid contamination.

This product and its components should be handled only by persons trained in laboratory techniques and used in accordance with the principles of good laboratory practice. All chemicals should be considered potentially hazardous; therefore, when handling chemical reagents, it is advisable that suitable protective clothing, such as laboratory overalls, safety glasses and gloves be worn. Care should be taken to avoid contact with skin or eyes. In case of contact with skin or eyes, wash immediately with water. See the appropriate Material Safety Data Sheet for specific recommendations.

GenomiPhi™ V2 Amplification Kit is optimized for whole genome amplification from at least 10 ng of high quality genomic DNA template. Use of less DNA or low quality DNA (such as degraded DNA, or DNA from formalin fixed paraffin embedded samples) can result in amplification bias.

No amplification product is produced in the absence of template DNA up to 1.5 hours amplification. However, if amplification reactions are carried out for more than 1.5 hours, they may produce some artifact DNA synthesis in no-template controls.

Wear gloves at all times during the preparation to avoid contamination.

2.2. Storage conditions

Store the kit at -70°C.

The enzyme mix must be stored at -70°C; all other components may be stored at -20°C. Thaw components on ice and maintain at 0°C to 4°C during handling.

2.3. Expiry

This product has been designed to deliver high quality results for up to 12 months from the manufacturing date. Please refer to the detail expiration date on the product label.

DOMINIQUE DUTSCHER SAS

3. Components

3.1 Kit Contents

Genomiphi V2 DNA Amplification Kit

Cat. No.	500 Reactions 25-6600-32	100 Reactions 25-6600-31	25 Reactions 25-6600-30
Sample Buffer (Green Cap)	5 x 0.9 ml	1 x 0.9 ml	1 x 225 µl
Reaction Buffer (Blue Cap)	5 x 0.9 ml	1 x 0.9 ml	1 x 225 µl
Enzyme Mix (Yellow Cap)	5 x 100 µl	1 x 100 µl	1 x 25 µl
Control DNA (Lambda), 10 ng/µl	1 x 20 µl	1 x 20 µl	1 x 20 µl

3.2. Reagents to be supplied by the user

Liquid-handling supplies - Vials, pipettes, micro centrifuge, and vacuum centrifuge. Perform all amplification reactions in plastic micro centrifuge tubes (typically 0.5 ml), or in 96-well or 384-well plates suitable for sealing and incubating at 30°C.

Thermocycler or water baths - for incubations at 30°C, 65°C and 95°C.

DOMINIQUE DUTSCHER SAS

4. Product description

4.1 The basic principle

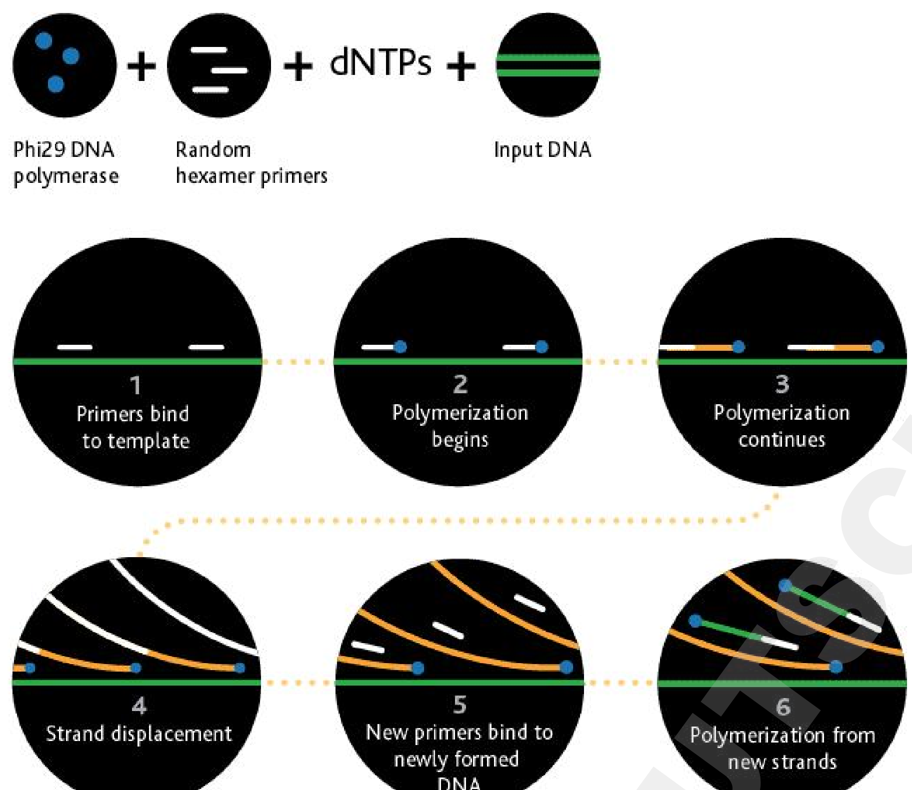


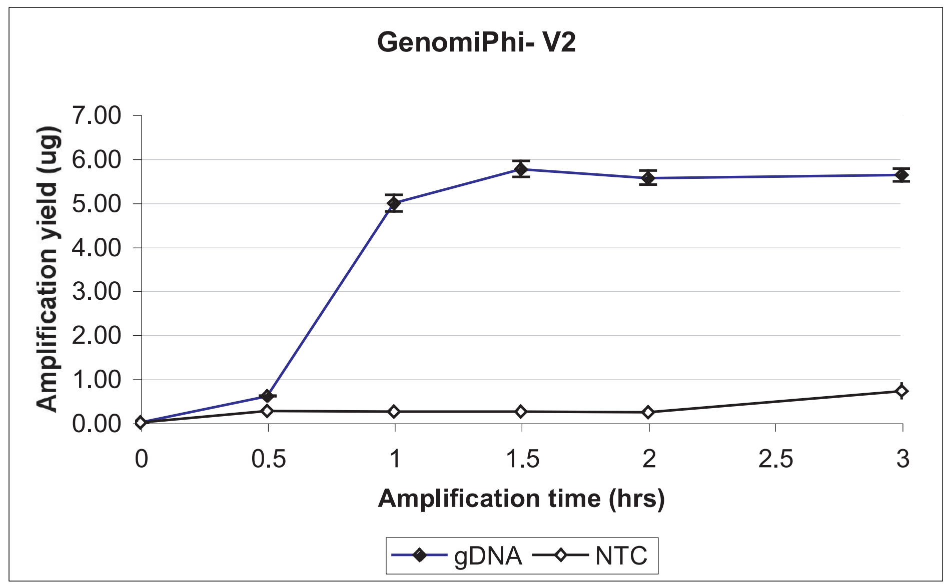
Figure 1 Overview of the **GenomiPhi V2 DNA Amplification Kit** procedure

Figure 1 shows an overview of whole genome amplification by isothermal stand displacement using the **GenomiPhi V2 DNA Amplification Kit**. DNA is briefly heat-denatured then cooled in sample buffer containing random hexamers that non-specifically bind to the DNA. A master-mix containing DNA polymerase, additional random hexamers, nucleotides, salts and buffers is added and isothermal amplification proceeds at 30°C for 1.5 hours. After amplification the enzyme is heat inactivated during 10 minute incubation at 65°C.

4.2 Kit specifications

Typical amplification kinetics with **GenomiPhi V2 DNA Amplification Kit** is shown in Figure 2 below. Microgram quantities of DNA are generated from nanogram amounts of starting material in 1.5 hours. Typical DNA yields from a **GenomiPhi V2 DNA Amplification Kit** reaction are 4–7 µg per 20 µl reaction when starting with 10 ng of purified DNA. Kinetics will vary if crude or un-quantified samples are amplified. Increased reaction times (2 hours) may be helpful for samples such as crude blood or buccal swabs. Reactions containing no DNA do not produce any product during 1.5 h reactions. The average product length is greater than 10 kb. DNA replication is extremely accurate due to the proofreading 3'– 5' exonuclease activity of the DNA polymerase (3, 4).

DOMINIQUE DUTSCHER SAS



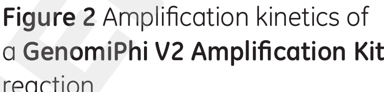


Figure 2 Amplification kinetics of a **GenomiPhi V2 Amplification Kit** reaction.

Figure 2 shows the comparison of amplification of 10 ng purified gDNA with no template control (NTC). Most commercial DNA isolation kits and homemade purification procedures produce suitable DNA for the amplification

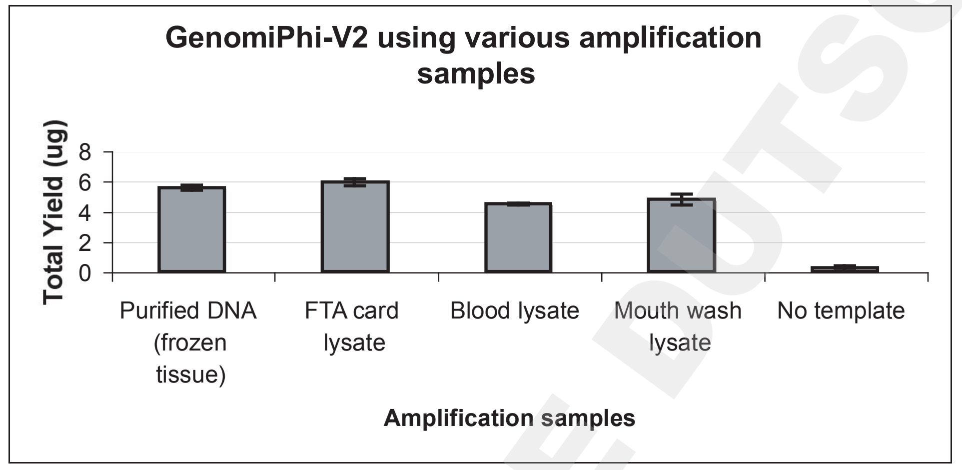


Figure 3 Amplification of various samples using **GenomiPhi V2 Amplification Kit.**

Figure 3 shows the amplification yields for **GenomiPhi V2 Amplification Kit** using purified DNA (10 ng) or non-purified cell lysates. Amplification protocols are described later in the booklet.

DOMINIQUE DUTSCHER SAS

5. Protocols

5.1. Short Protocol

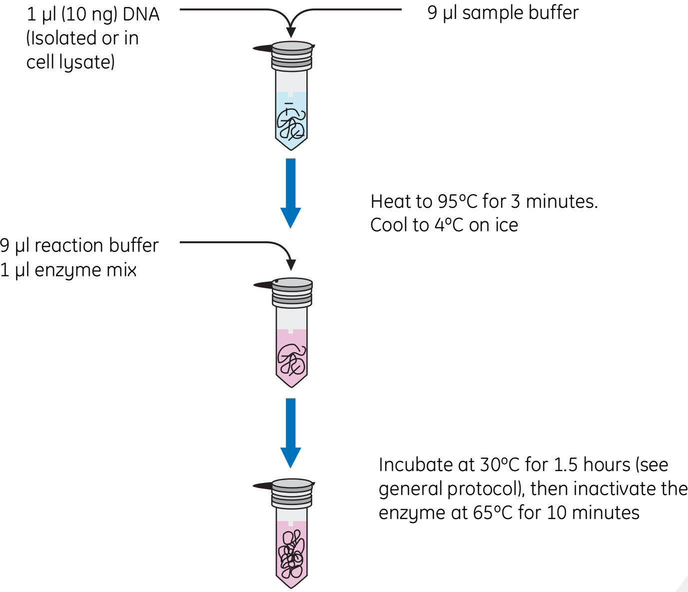


Figure 4 Schematic representation of **GenomiPhi V2 Amplification Kit** protocol.

5.2 General Protocol

The steps outlined below describe a general protocol for amplifying template DNA. This protocol should be considered a starting point for optimizing the reaction in your laboratory.

Step	
1. Mix sample buffer with template DNA **Add 9 µl** Sample Buffer to **1 µl** of 10 ng template DNA. Template DNA should be resuspended in TE or water.	9 µl Sample Buffer 1 µl DNA Template (10 ng)
2. Denature template DNA Heat the samples to **95°C** for **3 minutes** then cool to **4°C** on ice. Heating the DNA for longer than 3 minutes or at higher temperatures can cause damage to the DNA.	95°C for 3 minutes
3. Prepare the master mix for each amplification reaction For each amplification reaction, on ice combine **9 µl** of Reaction Buffer with **1 µl** of Enzyme Mix. Prepare the master mix only in sufficient quantities and immediately prior to use. Keep the master mix on ice and discard any unused portion. The master mix contains all the components required for DNA amplification and will generate amplification products if exposed to temperatures > 4°C for sufficient time.	9 µl Reaction Buffer 1 µl Enzyme Mix

DOMINIQUE DUTSCHER SAS

4. Transfer master mix to cooled sample. Add **10 µl** of prepared master mix from **Step 3** to each cooled sample, on ice.	10 µl Master Mix
5. Incubate for DNA amplification. Incubate the samples at **30°C** for **1.5 hours.**	30°C for 1.5 hours
6. Inactivate the Phi29 DNA polymerase enzyme. Heat the samples to **65°C** for **10 minutes** then cool to **4°C.** Heating is required to inactivate the exonuclease activity of the DNA polymerase which may otherwise begin to degrade the amplification product.	65°C for 10 minutes
7. Storage of amplified material Store amplification reactions at **-20°C.** GenomiPhi DNA V2 amplification products should be stored and treated as genomic DNA.	

5.3 Chemical denaturation and amplification of template DNA

The steps outlined below describe a general protocol for amplifying template DNA.

1. Prepare denaturation solution and neutralization buffer. **Denaturation Solution** 400 mM KOH 10 mM EDTA **Neutralization Buffer** 400 mM HCl 600 mM Tris-HCl, pH 7.5	
2. Denature template DNA Mix **1 µl** of template (10 ng) with **1 µl** of Denaturation Solution. Incubate at room temperature for 3 minutes. Mix by pipetting up and down. Do not vortex.	1 µl of DNA + 1 µl Denaturation Solution RT for 3 minutes
3. Neutralize DNA solution. Add **1 µl** of Neutralization Buffer and store on ice. Mix by pipetting up and down. Do not vortex.	1 µl Neutralization Buffer
4. Prepare the master mix for each amplification reaction For each amplification reaction, on ice combine **7 µl**	

DOMINIQUE DUTSCHER SAS

of Sample Buffer and **9 µl** of Reaction Buffer, with **1 µl** of Enzyme Mix.

Prepare the master mix only in sufficient quantities and immediately prior to use. Keep the master mix on ice and discard any unused portion. The master mix contains all the components required for DNA amplification and will generate amplification products if exposed to temperatures > 4°C for sufficient time.

5. **Transfer master mix to cooled sample.**

   Add **17 µl** of prepared master mix from **Step 4** to each cooled sample, on ice.

   17 µl Master Mix

6. **Incubate for DNA amplification.**

   Incubate the samples at 30°C for **1.5 hours.**

   30°C for 1.5 hours

7. **Phi29 DNA polymerase enzyme.**

   Heat the samples to **65°C** for **10 minutes** then cool to **4°C.**

   65°C for 10 minutes

   Heating is required to inactivate the exonuclease activity of the DNA polymerase which would otherwise begin to degrade the amplification product.

8. **Storage of the amplified material**

   Store amplification reactions at **-20°C.**

   GenomiPhi DNA V2 amplification products should be stored and treated as genomic DNA.

5.4 DNA amplification from blood cells

Note: Components of blood (e.g. heme) can be inhibitory to the GenomiPhi reaction. Amplification is faster and more reproducible when the blood sample contains relatively low blood components prepared by such as dilution. A reaction time of 2 hours is suggested.

1. **Prepare cell lysis solution and neutralization buffers.**

   Cell lysis solution
   400 mM KOH
   10 mM EDTA
   100 mM DTT

   Neutralization Buffer
   400 mM HCl
   600 mM Tris-HCl, pH 7.5

2. **Cell lysis**

   Dilute blood 3 times with physiological buffer, e.g., PBS. Add one part of this mixture with one part of cell lysis solution. Mix well with gentle tapping and incubate on ice for 10 minutes.

   1 µl of diluted blood cells
   +
   1 µl cell lysis solution
   10 minutes on ice

DOMINIQUE DUTSCHER SAS

Mix by pipetting up and down. Do not vortex.

---

3. **Neutralize cell lysate.**

   Add one part of neutralization buffer to the cell lysate, mix well and store on ice.

   1 µl Neutralization Buffer

   Mix by pipetting up and down. Do not vortex.

---

4. **Prepare the master mix for each amplification reaction**

   For each amplification reaction, on ice combine **7 µl** of Sample Buffer and **9 µl** of Reaction Buffer, with **1 µl** of Enzyme Mix.

   Prepare the master mix only in sufficient quantities and immediately prior to use. Keep the master mix on ice and discard any unused portion. The master mix contains all the components required for DNA amplification and will generate amplification products if exposed to temperatures > 4°C for sufficient time.

---

5. **Transfer master mix to cooled sample.**

   Add **17 µl** of prepared master mix from **Step 4** to each cooled sample, on ice.

   17 µl Master Mix

---

6. **Incubate for DNA amplification.**

   Incubate the samples at 30°C for **2 hours.**

   30°C for 2 hours

---

7. **Phi29 DNA polymerase enzyme.**

   Heat the samples to **65°C** for **10 minutes** then cool to **4°C.**

   65°C for 10 minutes

   Heating is required to inactivate the exonuclease activity of the DNA polymerase which would otherwise begin to degrade the amplification product.

---

8. **Storage of the amplified material**

   Store amplification reactions at **-20°C.**

   GenomiPhi DNA V2 amplification products should be stored and treated as genomic DNA.

5.5 DNA amplification from cheek cells (mouth wash or buccal swab)

---

1. **Prepare cell lysis solution and neutralization buffers.**

   Cell lysis solution
   400 mM KOH
   10 mM EDTA
   100 mM DTT

DOMINIQUE DUTSCHER SAS

Neutralization Buffer
400 mM HCl
600 mM Tris-HCl, pH 7.5

2. Cell lysis

Wash cheek cells (from mouth wash or buccal swab) with PBS and centrifuge. To the cell pallet add enough cell lysis solution (**10 µl**) to submerge the cells completely. Mix well with gentle tapping and incubate on ice for 10 minutes.

Palleted cheek cells
+
10 µl cell lysis solution
10 minutes on ice

Mix by pipetting up and down. Do not vortex.

3. Neutralize cell lysate.

Add one part of neutralization buffer **(10 µl**) to the cell lysate, mix well and store on ice.

10 µl Neutralization Buffer

Mix by pipetting up and down. Do not vortex.

4. Prepare the master mix for each amplification reaction

For each amplification reaction, on ice combine **9 µl** of Sample Buffer and **9 µ**l of Reaction Buffer, with **1 µl** of Enzyme Mix.

Prepare the master mix only in sufficient quantities and immediately prior to use. Keep the master mix on ice and discard any unused portion. The master mix contains all the components required for DNA amplification and will generate amplification products if exposed to temperatures > 4°C for sufficient time.

5. Transfer master mix to 1 µl of cooled cell lysate.

Add **19 µl** of prepared master mix from **Step 4** to **1 µl** of neutralized cell lysate on ice.

19 µl
Master Mix

6. Incubate for DNA amplification.

Incubate the samples at **30°C** for **2 hours.**

30°C
for 2 hours

7. Phi29 DNA polymerase enzyme.

Heat the samples to **65°C** for **10 minutes** then cool to **4°C.**

65°C
for 10 minutes

Heating is required to inactivate the exonuclease activity of the DNA polymerase which would otherwise begin to degrade the amplification product.

8. Storage of the amplified material Store amplification reactions at **-20°C.**

GenomiPhi DNA V2 amplification products should be stored and treated as genomic DNA.

DOMINIQUE DUTSCHER SAS

5.6 DNA amplification from blood or buccal swab-blotted paper (e.g., Whatman's FTA paper or Guthrie card)

1. Prepare cell lysis solution and neutralization buffers.

Cell lysis solution
400 mM KOH
10 mM EDTA
100 mM DTT

Neutralization Buffer
400 mM HCl
600 mM Tris-HCl, pH 7.5

2. Cell lysis

Cut the blood-blotted FTA paper in small pieces such as 3 mm x 3 mm square or punch out a 3 mm diameter circle from it, and wash the blotted FTA card piece with FTA purification reagent (Whatman catalog number WB120204) in a micro centrifuge tube. Add enough **(10 µl)** cell lysis solution to submerge the paper completely. Mix well with gentle tapping and incubate on ice for **10 minutes.**

Washed FTA card piece blotted with blood or cheek cells + 10 µl cell lysis solution

10 minutes on ice

Mix by pipetting up and down. Do not vortex.

3. Neutralize cell lysate.

Add one part of neutralization buffer **(10 µl)** to the cell lysate, mix well and store on ice.

10 µl Neutralization Buffer

Mix by pipetting up and down. Do not vortex.

4. Prepare the master mix for each amplification reaction

For each amplification reaction, on ice combine **9 µl** of Sample Buffer and **9 µl** of Reaction Buffer, with **1 µl** of Enzyme Mix.

Prepare the master mix only in sufficient quantities and immediately prior to use. Keep the master mix on ice and discard any unused portion. The master mix contains all the components required for DNA amplification and will generate amplification products if exposed to temperatures > 4°C for sufficient time.

5. Transfer master mix to 1 µl cooled cell lysate.

Add **19 µl** of prepared master mix from **Step 4** to **1 µl** of neutralized cell lysate on ice.

19 µl Master Mix

6. Incubate for DNA amplification.

Incubate the samples at **30°C** for **2 hours.**

30 °C for 2 hours

DOMINIQUE DUTSCHER SAS

5. **Phi29 DNA polymerase enzyme.**

   Heat the samples to **65°C** for 10 minutes then cool to **4°C.**

   Heating is required to inactivate the exonuclease activity of the DNA polymerase which would otherwise begin to degrade the amplification product.

   65°C
   for 10 minutes

6. **Storage of the amplified material**

   Store amplification reactions at -**20°C.**

   GenomiPhi DNA V2 amplification products should be stored and treated as genomic DNA.

5.7 Quantification of amplification products

Quantification is generally not required as every reaction will yield approximately the same amount of DNA. **Quant-iT™ PicoGreen® dsDNA quantification reagent** (Invitrogen, P7581) is recommended if accurate quantitation is required.

Quantification of non-purified amplification products by UV absorption will generate inaccurate results due to the presence of unused hexamers in the completed reaction.

1. **Prepare TE buffer.**

   Dilute the concentrated 20 x TE buffer included in the kit to 1 x concentration using water.

   Prepare 1 x TE buffer

   Use only sterile, distilled, DNase free water when preparing the dilution to ensure accurate quantification.

2. **Prepare 1:25 dilution of PicoGreen reagent.**

   Determine the required volume of a 1:25 dilution of PicoGreen reagent.

   • Volume = 100 µl/sample x # of samples

   Determine the volume of stock PicoGreen reagent necessary to produce the required volume of a 1:25 dilution

   • Volume = $\frac{\text{volume of required dilution}}{25}$

   Reagent adsorbs to glass surfaces. Use plastic ware only. Protect the solution from light at all times.

3. **Prepare the λ DNA standard curve. Dilute** the λ DNA standard supplied in the Quant-iT PicoGreen® kit to a **10 ng/µl** working solution. Use this working stock to prepare a standard curve (see below).

   Add 100 µl of each dilution to each well of the assay plate.

DOMINIQUE DUTSCHER SAS

Standard Number	λ DNA (10(ng)	λ DNA (10 ng/µl)	1 x TE
1	600	60 µl	40 µl
2	500	50 µl	50 µl
3	400	40 µl	60 µl
4	200	20 µl	80 µl
5	100	10 µl	90 µl
6	50	5 µl	95 µl
7	25	2.5 µl	97.5 µl
8	0	0 µl	100 µl

4. Dilute the GenomiPhi V2 amplification products.

Dilute the GenomiPhi V2 amplification products 1:10 by **adding 180 µl** of 1 x TE to each amplification reaction. Due to the viscosity of the amplification product, mix amplification products thoroughly by vortexing heavily.

Dilute 1:10

5. Add diluted GenomiPhi amplification products to the assay plate.

Aliquot **90 µl** of 1 x TE into each sample well. **Add 10 µl** of diluted sample for a final volume of **100 µl.**

90 µl of TE
+
10 µl of diluted sample

Because the amplification product is diluted **before** the assay, the dilution factor must be taken into consideration when calculating total yields.

6. Add diluted PicoGreen to sample wells.

Add 100 µl of the 1:25 dilution of PicoGreen to all wells containing standards and samples. Mix contents well by pipetting up and down.

Seal the plate with foil and spin in micro plate centrifuge for 1 minute at < 200 x *g* to eliminate bubbles.

Protect plate from light at all times. The plate must be read 5-10 minutes after addition of PicoGreen reagent to ensure accurate quantification

7. Measure the sample fluorescence.

Place the sample assay plate into a fluorescence micro plate reader.

Set the fluorescence reader at the following parameters:

- Excitation wavelength: **480 nm**
- Emission wavelength: **520 nm**
- Gain: **Optimal**

If it is not possible to set the instrument gain to optimal, find a way to have the instrument read the sample where the highest DNA concentration generates readings that fall within the linear dynamic range of the

DOMINIQUE DUTSCHER SAS

instrument.

8. Calculate the concentration of the amplification product.

Generate a standard curve of fluorescence versus DNA concentration. Determine the concentration of GenomiPhi V2 Kit amplified products from the equation of the line derived from the standard curve.

DOMINIQUE DUTSCHER SAS

6. Appendix

6.1 Troubleshooting

Problem	Possible cause and suggestions
Reduced yield/ no amplification product	**Contamination of template DNA** • Excessive contaminants carried over from the starting material can inhibit the DNA polymerase. Dilute or clean-up the DNA and re-am plify. • Extending the amplification time will help when inhibitory material is causing reduced yields. **Inactive Enzyme** • It is critical that the enzyme be stored properly. The **Enzyme Mix** should be stored at -**70°C.** If the material will be consumed within **2 months, -20°C** storage may be used. The freezer must **not** be a frost-free unit. • Perform a control reaction to confirm performance of the enzyme. **Low quality DNA** • Amplification kinetics strongly favors intact templates. Avoid template preparation steps that can damage DNA. **Prolonged denaturation** • Heating at **95°C** for 3 minutes is sufficient to denature template DNA and facilitate primer annealing. Longer denaturing times can nick the template and decrease the amplification efficiency.
Poor performance in downstream applications	**Degraded/low amounts of template DNA** • In the absence of input DNA or poor quality of input DNA, there will be no or minimal DNA synthesis in the amplification reactions within 2 hours. • Degraded or low amounts of starting DNA template may not amplify consistently or representatively. Increase the amount of starting DNA. • Use high quality genomic DNA for amplification. **Inhibition of optimized downstream conditions** • Starting material components can inhibit amplification reaction. Purify the starting material using a suitable column prior to amplification. • For some downstream applications, components of the GenomiPhi V2 reaction will alter previously optimized downstream conditions. Purify the amplification products using a suitable column after amplification.

DOMINIQUE DUTSCHER SAS

6.2 References

1. Dean, F. *et al., Genome Research* **11**, 1095–1099 (2001).
2. Lizardi, P. *et al., Nat. Genet*. **19**, 225–232 (1998).
3. Estaban, J.A. *et al., J. Biol. Chem.* **268**, 2719–2726 (1993).
4. Nelson, J.R. *et al; BioTechniques* **32**, S44-S47 (2002).

6.3 Related Products

DNA Purification Products

GFX™ Genomic Blood DNA Purification Kit

Nucleon™ BACC Genomic DNA Extraction Kits

GFX™ Micro Plasmid Prep Kit

MicroSpin™ Columns

GFX™ PCR DNA and Gel Band Purification Kit

CyScribe™ GFX™ Purification Kit

ProbeQuant™ G-50 Micro Columns

PCR Products

PureTaq™ Ready-To-Go™ PCR Beads

FideliTaq™ PCR Master Mix

dNTP Set, 100 mM Solution (dATP, dGTP, dCTP, dTTP)

Exo-SAP IT

Taq Polymerase™

Sequencing Products

MegaBACE™ DNA Analysis System

DYEnamic™ ET Dye Terminator Cycle Sequencing Kit

Autoseq™ G-50

DOMINIQUE DUTSCHER SAS

DOMINIQUE DUTSCHER SAS

GE Healthcare offices:

GE Healthcare Bio-Sciences AB
Björkgatan 30 751 84
Uppsala Sweden

GE Healthcare Europe GmbH
Munzinger Strasse 5 D-79111
Freiburg Germany

GE Healthcare UK Limited
Amersham Place
Little Chalfont Buckinghamshire
HP7 9NA UK

GE Healthcare Bio-Sciences Corp
800 Centennial Avenue
P.O. Box 1327
Piscataway
NJ 08855-1327 USA

GE Healthcare Bio-Sciences KK
Sanken Bldg. 3-25-1
Hyakunincho Shinjuku-ku
Tokyo 169-0073
Japan

GE Healthcare regional office contact numbers:

Asia Pacific
Tel: +65 6275 1830
Fax: +65 6275 1829

Australasia
Tel: + 61 2 9899 0999
Fax: +61 2 9899 7511

Austria
Tel: 01/57606-1619
Fax: 01/57606-1627

Belgium
Tel: 0800 73 888
Fax: 02 416 82 06

Canada
Tel: 1 800 463 5800
Fax: 1 800 567 1008

Central, East, & South East Europe
Tel: +43 1 982 3826
Fax: +43 1 985 8327

Denmark
Tel: 45 16 2400
Fax: 45 16 2424

Eire
Tel: 01494 544000
Fax: 0044 1494 542010

Finland & Baltics
Tel: +358-(0)9-512 39 40
Fax: +358 (0)9 512 39 439

France
Tel: 01 6935 6700
Fax: 01 6941 9677

Germany
Tel: (089) 96281 660
Fax: (089) 96281 620

Greater China
Tel: +852 2100 6300
Fax: +852 2100 6338

Italy
Tel: 02 27322 1
Fax: 02 27302 212

Japan
Tel: +81 3 5331 9336
Fax: +81 3 5331 9370

Latin America
Tel: +55 11 3933 7300
Fax: + 55 11 3933 7304

Middle East & Africa
Tel: +30 210 9600 687
Fax: +30 210 9600 693

Netherlands
Tel: 0800 8282821
Fax: 0800 8282824

Norway
Tel: 815 65 555
Fax: 815 65 666

Portugal
Tel: 21 417 7035
Fax: 21 417 3184

Russia & other C.I.S. & N.I.S
Tel: +7 495 956 5177
Fax: +7 495 956 5176

Spain
Tel: 93 594 49 50
Fax: 93 594 49 55

Sweden
Tel: 018 612 1900
Fax: 018 612 1910

Switzerland
Tel: 0848 8028 12
Fax: 0848 8028 13

UK
Tel: 0800 616928
Fax: 0800 616927

USA
Tel: +1 800 526 3593
Fax: +1 877 295 8102

DOMINIQUE DUTSCHER SAS

http://www.gehealthcare.com/lifesciences
GE Healthcare UK Limited
Amersham Place, Little Chalfont,
Buckinghamshire,
HP7 9NA UK

25-6600-30WP Rev B 2006