Purify. Amplify. Simplify.

# Fast and simple DNA sample preparation

illustra GenomiPhi illustra TempliPhi



# Simplify the process of DNA isolation with illustra GenomiPhi & TempliPhi

The Phi29 DNA polymerase–based, isothermal DNA amplification method is a simple, reliable alternative to traditional DNA isolation procedures. It produces high-quality DNA from small amounts of starting material and simplifies the DNA preparation process. The method is easy to automate and the DNA yield is consistent from sample to sample. The output DNA is ready for direct use in downstream analyses such as sequencing and genotyping.

# Phi29 DNA polymerase-based kits

GenomiPhi™ V2 DNA Amplification Kit GenomiPhi HY DNA Amplification Kit TempliPhi™ Large Construct DNA Amplification Kit TempliPhi HT DNA Amplification Kit TempliPhi 100/500 DNA Amplification Kit TempliPhi Sequence Resolver Kit





# Comparison of different whole genome amplification methods

The most commonly used WGA methods are Phi29 DNA polymerase based and polymerase chain reaction (PCR) based, including degenerate-oligonucleotide primed PCR (DOP-PCR) and primer extension preamplification (PEP) PCR. PCR-based protocols can be complex, the amplified DNA fragment is relatively short, and amplified PCR products are not always representative of the original genomic DNA. With illustra<sup>™</sup> GenomiPhi kits, both the fidelity and processivity of Phi29 DNA Polymerase surpass that of *Taq* polymerase, maximizing your ability to obtain representative DNA in a simple, isothermal reaction for various DNA analyses.





# Phi29 DNA polymerase generates high molecular weight products...

Purified human genomic DNA (10 ng) was amplified with each of the three kits per standard protocol. Amplified samples were separated by pulsed-field gel electrophoresis (1% gel, 48 h). The gel was stained with ethidium bromide and then scanned on a Typhoon™ scanner.



### ...and high-quality representative DNA

10 na of purified human aenomic DNA was amplified with each of the three kits per standard protocol. They were loaded on a 1% pulsed field gel electrophoresis for 48 hours and then stained with ethidium bromide. Gel was scanned on Typhoon scanner. Representative amplification with illustra GenomiPhi DNA Amplification Kit. Human genomic DNA samples were amplified in triplicate using various DNA amplification methods, followed by genotyping for over 5000 SNPs using the Linkage Panel IV chip performed by Illumina<sup>™</sup> Genotyping Service. Amplified DNA produced with illustra GenomiPhi DNA Amplification Kit was 100% concordant with genomic DNA.

# illustra GenomiPhi

# Easy method for genomic DNA preparation

Genomic DNA isolation is a fundamental step in genetic analysis, and obtaining high-quality DNA is vital for success. GE Healthcare's illustra GenomiPhi DNA amplification kits provide an easy way to prepare genomic DNA. The kits enable you to prepare sufficient DNA for genetic analysis from biological materials of limited availability and simplify simultaneous preparation of multiple samples. High yield genomic DNA means that you minimize re-isolation of DNA from the original source material, thereby saving you time and money.

Since the launch of the first GenomiPhi DNA amplification kit in 2003, hundreds of researchers have validated this method and cited the use of this kit in over one hundred publications by 2006. It delivers highly representative and reliable whole genome amplification (WGA).

# illustra GenomiPhi HY DNA Amplification Kit

- Midi-scale genomic DNA preparation: 40 to 50 µg in 4 h
- Less hands-on time compared to traditional isolation methods
- Simple, automation-friendly protocol with great reproducibility

# illustra GenomiPhi V2 DNA Amplification Kit

- Quick mini-scale genomic DNA preparation: 4 to 7  $\mu$ g in 2 h
- One simple protocol for all different types of source material
- No background amplification in negative controls

# Phi29 DNA polymerase–based DNA amplification workflow versus traditional DNA purification workflow



Pure genomic DNA

# illustra GenomiPhi V2 DNA Amplification Kit

illustra GenomiPhi V2 DNA Amplification Kit is designed for quick preparation of 4 to 7  $\mu$ g of genomic DNA from different types of samples for your PCR, genotyping, cloning applications, and other research needs.

With a 2 h reaction time, the illustra GenomiPhi V2 kit enables you to prepare the DNA and complete your downstream application all within the same day. The no-DNA control reaction will not generate any background amplification. It will provide you a clear indication of which of your sample preparations really worked, thus saving you from spending time and money processing nonspecific amplification products..

With the ability to generate µg amounts of highly representative genomic DNA from ng amounts of starting material, illustra GenomiPhi V2 DNA Amplfication Kit can stretch limited genomic DNA stocks into a nearly unlimited supply.

# Ordering information

illustra GenomiPhi								
Product	Pack size	Code number						
illustra GenomiPhi V2 DNA Amplification Kit	25 rxns	25-6600-30						
illustra GenomiPhi V2 DNA Amplification Kit	100 rxns	25-6600-31						
illustra GenomiPhi V2 DNA Amplification Kit	500 rxns	25-6600-32						



<b>A</b> −A	5.3	5.5	5.5	5.8	5.8	5.2	6	5.9	5.9	5.9	•
VO →	5.3	5.2	5.1	5.1	5.3	5.4	5.4	5.1	5.3	5	ATOR 1
	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	OPER/
i≥ ↓	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	₩
A-A	5.4	5.6	5.6	5.3	5.4	5.6	5.6	5.8	5.4	5.4	•
J □ -	5.7	5.9	5.8	5.5	5.4	5.9	5.9	5.7	5.3	5.3	ATOR 2
TC-	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	OPER
₹	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	0.1	↓

### Consistent yield with no background amplification

Multiwell assay with illustra GenomiPhi V2 DNA Amplification Kit NTC: no template control; DNA: 10 ng purified genomic DNA was used as starting material. (units =  $\mu$ g)



# SNP analysis of amplified products using Affymetrix 10k human genome chip

Individual human genomic DNA (gDNA) obtained from Coriell was amplified with illustra GenomiPhi V2 kits and subjected to analysis on Affymetrix™ 10k SNP chip

(green = % call rate; blue = % concordance).



### illustra GenomiPhi V2 using various samples

Genomic DNA preparation using illustra GenomiPhi V2 kit from various source materials.

# illustra GenomiPhi HY DNA Amplification Kit

illustra GenomiPhi HY DNA Amplification Kit is designed to meet your needs for high-yield genomic DNA preparation for applications such as SNP genotyping, STR whole genome scan, array CGH, and DNA archiving. Compared to traditional isolation methods, illustra GenomiPhi HY DNA Amplification Kit allows you to start with small amounts of sample and generate 40 to 50 µg in about 4 h. The method is very simple and robust. It reduces your hands-on time for simultaneous handling of multiple samples and generates consistent results.

If you already have 10 ng or more of purified genomic DNA from your sample, illustra GenomiPhi HY DNA Amplification Kit can also help you to produce more of the same DNA quickly. You do not need to go back to the original source material; thus you save time and money. This will also allow you to share your precious DNA sample more easily with your collaborators or colleagues. illustra GenomiPhi HY DNA Amplification Kit produces high-quality DNA to meet your downstream application needs.

# And and a second descent des

# Ordering information

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Product	Pack size	Code number	
illustra GenomiPhi HY DNA Amplification Kit	100 rxns	25-6600-20	
illustra GenomiPhi HY DNA Amplification Kit	1000 rxns	25-6600-25	

illustra ConomiDhi kit	provides bigh	uiold and re	nraducibilitu
mustru Gerionnenn kit	provides night	yleia ana re	producibility

Two-operator multiwell assay with illustra GenomiPhi HY DNA Amplification Kit. (units = µg)

										-
46.3	48.5	48.1	46.9	48.4	45.9	41.1	47.2	48.5	39.1	•
45.1	47.5	47.2	47.2	47.2	47.2	46.7	51	47.9	46.7	ATOR 1
44.7	45.9	46.3	45.4	47.6	48.5	41.1	47	46.4	47.6	OPER
44	43.9	45.4	45	45.1	45.4	44.4	45.1	43.4	44.3	↓
49.7	47.7	48.7	46.7	52.8	43.9	45.1	49	48.1	48	1
49.1	47.4	48.4	47.8	45.9	45.1	39.7	44.6	46.1	50.5	ATOR 2
46.1	42.3	47.6	46.3	46.6	45.2	43.2	46.5	42.3	47.1	OPER
46	45.6	45.6	43.8	42.7	42.7	42.4	38.9	42	42.1	↓



# High quality DNA provide accurate genotyping calls

Genomic DNA and amplified samples were genotyped for 15 SNPs in a multiplex panel using SEQUENOM™ iPLEX™ chemistry. All 18 amplified samples were automatically called and gave the correct genotypes compared to genomic DNA. Manual calling was required for 1.2% of genomic DNA samples.

Representative data for two SNPs is shown. Data kindly provided by Dr. Nadine Norton, Department of Psychological Medicine, Cardiff University, Cardiff, Wales, UK.

# illustra TempliPhi DNA Amplification Kits

# **Culture-free DNA preparation**

illustra TempliPhi generated DNA can be used directly for sequencing and library construction, without further purification. These kits enable you to prepare circular DNA directly from bacterial colonies or glycerol stocks, eliminating the need for overnight culture. It reproducibly generates highquality DNA, with easily automated protocols allowing time and cost savings.

# illustra TempliPhi Large Construct DNA Amplification Kit

- For large DNA vector (e.g. fosmid and BAC) preparation
- Simple, automation-friendly protocol with great reproducibility

# illustra TempliPhi HT DNA Amplification Kit

- For high-throughput (> 1000 reactions/wk) sequencing template preparation
- Premixed formulation format to further reduce hands-on time
- Automation-friendly protocol with consistent DNA yield

# illustra TempliPhi 100/500 DNA Amplification Kit

- For low-to medium-throughput, small DNA vector preparation (e.g. pUC and M13)
- High-quality DNA in only 4 h
- Convenient and easy protocol
- Separate components for longer shelf life

# Ordering information

## illustra TempliPhi

Product illustra TempliPhi Large	Pack size	<b>Code number</b> 25-6400-80
Construct DNA Amplification Kit		
illustra TempliPhi 100/500 DNA Amplification Kit	100 rxns	25-6400-10
illustra TempliPhi 100/500 DNA Amplification Kit	500 rxns	25-6400-50
illustra TempliPhi DNA Sequencing HT DNA Amplification Kit	10000 rxns	25-6400-20





# Amplification kinetics of illustra TempliPhi kits

illustra TempliPhi HT DNA Amplification Kit generates approximately 4 µg of DNA in a 20-µl reaction in 16 h. illustra TempliPhi 100/500 kit generates approximately 2 µg of DNA in a 10-µl reaction in 4 h. illustra TempliPhi Large Construct Kit generates approximately 7 µg of DNA in a 20-µl reaction in 6 h.



### High-quality DNA generates accurate sequencing results Plasmid DNA template was amplified with the ilustra TempliPhi 100 Amplification Kit and subsequently sequenced using DYEnamic™ ET Terminator Cycle Sequencing Kit and analyzed on an ABI™ PRISM™ 3100 Genetic Analyzer.

# illustra TempliPhi Sequence Resolver Kit

# Difficult-template sequencing made easy

The illustra TempliPhi Sequence Resolver Kit is the latest DNA amplification technology product from GE Healthcare. The kit prepares circular DNA templates, such as plasmids, fosmids, and bacterial artificial chromosomes (BAC), and resolves sequencing stops caused by dinucleotide repeats and secondary structures to enable the generation of high-quality sequencing data from difficult-to-sequence templates. Compatible with a variety of sequencing chemistries, including DYEnamic ET terminator and BigDye™ v3.1 terminator cycle sequencing kits, the illustra TempliPhi Sequence Resolver Kit can be easily deployed upstream of standard sequencing protocols and instruments.

# Simple

- Resolution with a single kit—Solve the most common sequencing problems (repeats, sequencing stops, compressions) with one kit rather than multiple methods and reagents
- Culture-less preparation—Amplify DNA from bacterial colonies, glycerol stock, or purified DNA, saving costs in time, labor, and reagents

# **High Quality**

- More complete, accurate sequences—Improve difficult template sequencing success with up to 820 bp Phred 20 read lengths
- No purification required—Use prepared DNA directly in sequencing reactions, eliminating purification steps to achieve higher sequencing throughput

# Fast

• Just 15 min total hands-on time—Implement a simplified protocol with fewer steps and no overnight culturing

# Ordering information

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Product illustra TempliPhi Sequence Resolver Kit	Pack size 20 rxns	Code number 28-9035-29
illustra TempliPhi Sequence Resolver Kit	50 rxns	28-9035-30
illustra TempliPhi Sequence Resolver Kit	200 rxns	28-9035-31



# illustra TempliPhi Sequence Resolver Kit performance with different types of difficult templates

Difficult template type	Kit performance
Large (>20 bp) polynucleotide repeat	
Poly G	+
Poly C	+
Poly A	-
Poly T	-
Dinucleotide repeats	
AC/CA	++
AG/GA	++
AT/TA	-
CG/GC	++
СТ/ТС	++
GT/TG	++
Other types of difficult templates	
Secondary structure resulting in sequencing	g stop ++
GC rich templates	++
AT rich templates	-
Key: ++ kit usually helps resolve these types of sequence + kit sometimes helps resolve these types of prob	cing problems blems

- kit usually does not help resolve these types of sequencing problems

### Success of various difficult-to-resolve sequence types with traditional resolution methods and the illustra TempliPhi Sequence Resolver Kit

il Motif <sup>1</sup>	lustra TempliPhi Sequence Resolver Kit	None	DMSO (5%)	SequenceRx Enhancer A	Betaine (1 M)
GA dinuc repeat	731	251	590	307	293
GC rich	716	403	588	612	532
Poly G	480	378	379	410	413
TG dinuc repeat	829	374	379	388	386
Poly C	270	223	227	228	235
CA dinuc repeat/poly	/C 533	439	424	456	453
TC	829	40	7	31	47
TC at end of sequenc	e <b>746</b>	353	473	460	515
Inverted repeat	565	199	204	191	201
GC rich	550	291	489	350	366
Inverted repeat	689	481	588	702	764

<sup>1</sup> Plasmid templates were amplified using the illustra TempliPhi 100 DNA Amplification Kit or the illustra TempliPhi Sequence Resolver Kit. Amplified product (400 ng) was sequenced with the DYEnamic ET Terminator Kit per standard protocol. The indicated reagents were added to the sequencing reactions for the TempliPhi 100 amplified products. Sequencing reactions were analyzed on an ABI 3730xl DNA Analyzer, and Phred 20 read lengths (bp) were tabulated for all samples. The highlighted samples are those that yielded the longest read length for each motif.



### Comparative resolution of GC-rich region with dGTP BigDye v3.0 and the illustra TempliPhi Sequence Resolver Kit.

Plasmid template (1 µl) was amplified using either the illustra TempliPhi 100 Amplification Kit or the illustra TempliPhi Sequence Resolver Kit. Amplified product (400 ng) was sequenced with either the dGTP BigDye kit (illustra TempliPhi 100 product) or the DYEnamic ET Terminator kit (TempliPhi Sequence Resolver product) per the standard protocols. Sequencing reactions were analyzed on an ABI 3730xl DNA Analyzer. Note the GC-rich region toward the end of the sequence is poorly resolved with the dGTP technique, resulting in miscalling. The illustra TempliPhi Sequence Resolver Kit resolves compressions present with the dGTP technique. GE, imagination at work, and GE monogram are trademarks of General Electric Company.

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