Ovation® WGA System

The Only Linear WGA Technology for Uniform Representation and Quantitative Analysis of the Genome

Highlights of Ovation WGA System

- **Linear amplification for any downstream quantitative analysis:** Leveraging the power of the SPIA® technology generating µg amounts of amplified SPIA product for accurate, uniform representation of the whole genome from 10 ng of genomic DNA
- **Robust and consistent performance without the need for assay optimization:** Out-of-the-box experience reducing concerns for sample dropout, allele dropout or operator variation

Introduction

A broad range of genetic analysis tools is now available for detailed molecular characterization of genomic DNA (gDNA) samples, from q-PCR and microarrays to next-generation sequencing, with even more advanced techniques continuing to emerge. Applying these kinds of technologies to the analysis of limited clinical samples requires whole genome amplification methods that provide uniform representation and linearity across the whole genome and produce robust, fast and consistent results. Whole genome amplification is particularly critical to researchers in the areas of cancer, stem cell biology, neurobiology and immunology requiring efficient, large-scale whole genome genetic variation analysis, even on the smallest of samples.

Now there is a linear amplification method that uniformly represents the genome suitable for use in any quantitative downstream applications, the Ovation WGA System, a whole genome amplification reagent kit.

Because of the simplicity and robustness of the Ovation WGA System, consistent and reproducible performance can be obtained with minimal assay optimization leading to minimal allele or sample drop-outs. The combination of linear amplification and robustness offers scientists a new level of flexibility to generate a reliable, renewable source of faithfully amplified DNA from limited patient genomic DNA samples for analysis, today and in the future.

NuGEN®
imagine more from less™

Linear WGA Technology

Central to the Ovation WGA System is Single Primer Isothermal Amplification (SPIA) technology. Simple yet robust, the Ovation WGA System provides all necessary reagents to generate µg amounts of amplified SPIA product from 10 ng of genomic DNA in less than four hrs **(Figure 1)**.

A variety of samples can be used successfully with the Ovation WGA System, such as genomic DNA purified from cell lines, blood or tissue **(Figure 2A)**. The size of the resulting SPIA products ranges from 50 to 1,500 bases, suitable for a wide range of downstream analysis **(Figure 2B)**.

FIGURE 1 **Ovation WGA System Assay Diagram**

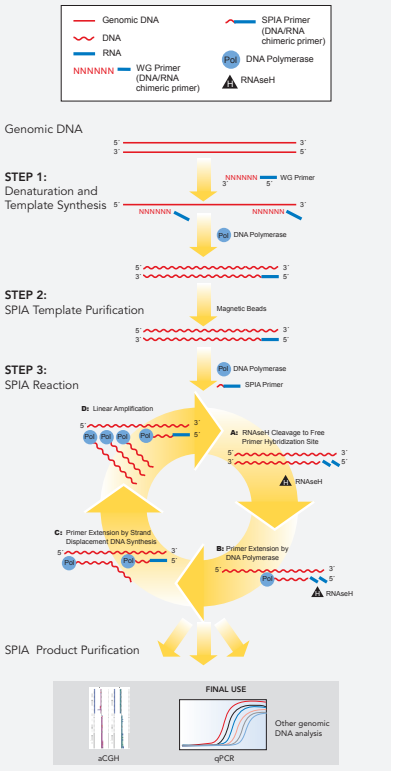
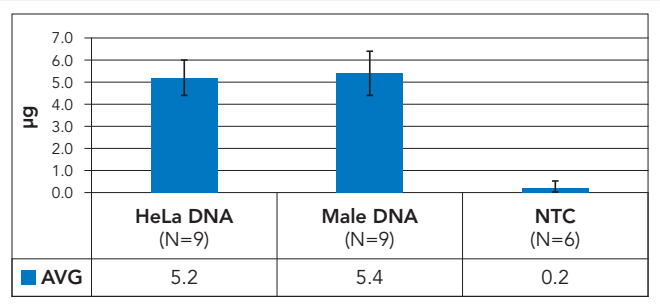
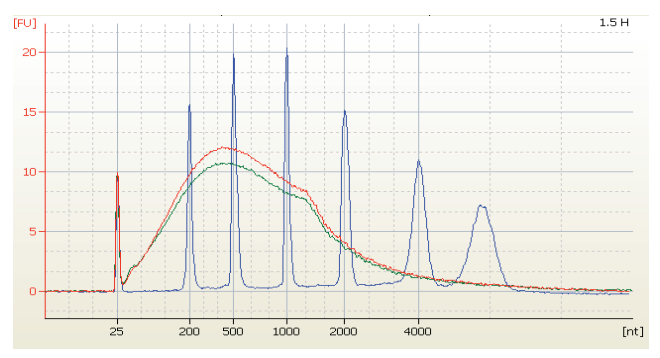


FIGURE 2A **Consistent Amplification of Genomic DNA**



Average yield and standard deviation are shown from replicate amplification reactions starting with 10 ng of genomic DNA isolated from a variety of sources, including cell lines (HeLa DNA obtained from NEB) and blood (male DNA obtained from Promega). Negligible amounts of DNA were obtained in the negative control sample (NTC).

FIGURE 2B **Size Distribution of SPIA products on BioAnalyzer RNA NanoChip**

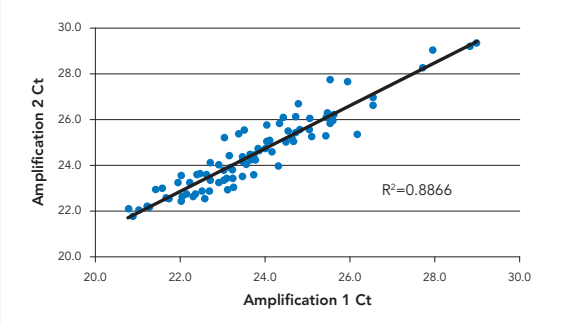


Consistent Performance Out of the Box

Most genetic research requires an even representation across the genome with minimal bias, regardless of the location of the allele of interest. This is particularly true for biomarker research. An ideal WGA technology would be applied to any allele without prior assay optimization, expediting the discovery and validation process.

The Ovation WGA System offers reproducibility and consistent performance across the entire genome. As shown in **Figure 3**, 87 q-PCR markers were designed to be evenly spaced across the human genome with approximately 5 loci per chromosome. These markers were selected blind to any Ovation WGA data, representing a naïve selection of alleles. Replicate amplification was performed and highly reproducible Ct values were obtained with no allele or sample dropout in the first attempt.

FIGURE 3 **Consistent Performance Across 87 Loci with No Allele Dropouts**



Genomic DNA was isolated from cultured cells with QIAGEN DNeasy Blood & Tissue Kit, and 10 ng was used in each triplicate amplification reaction with Ovation WGA System. Following amplification, 10 ng of SPIA product was used in each TaqMan assay. Average Ct values from TaqMan assays were plotted, comparing two independent amplification reactions with R^2 values between 0.85 and 0.89 for this set of markers. Note that zero allele dropouts were observed.

A Broad Range of Downstream Quantitative Applications

The Ovation WGA System provides accurate quantitation, suitable for clinically relevant copy number changes requiring detection of fold changes as low as 0.5 fold. **Figure 4** shows highly reproducible q-PCR results using trisomy samples compared with control samples as demonstrated as delta-Ct values. With no prior optimization, the q-PCR trisomy marker is able to quantitatively distinguish the trisomy samples from normal samples with consistency comparable to the unamplified samples.

FIGURE 4 **Robust and Reproducible Detection of Trisomy 21**

Six patient and six control blood samples were collected, and genomic DNA was isolated. Amplifications were performed in triplicate using 10 ng of isolated gDNA. TaqMan assays were carried out using 40 ng of unamplified gDNA or 40 ng of amplified SPIA product for each replicate. Delta-Ct values were derived from the relative comparison of a Chromosome 21 locus to a Chromosome 12 locus (GAPDH). In order to distinguish the additional Chr21 marker, the ΔΔCt method was employed, where the expected value from a 3:2 chromosomal imbalance should be 0.6. ΔΔCt values of 0.6 were consistently obtained from the SPIA amplified material and clearly distinguished the affected samples from the control samples, performing in a very similar way to the unamplified material.

Another powerful whole genome copy number analysis technology is Array Comparative Genome Hybridization (aCGH), which has been used broadly to survey the entire genome at high resolution in order to identify aberrant copy number changes. Ten ng of Genomic DNA from HeLa and normal male was amplified with Ovation WGA System. The SPIA product was then purified and labeled with Invitrogen's BioPrime Total for Agilent aCGH before hybridizing to Agilent 4x44 HD CGH Arrays. As shown in **Figure 5**, comparable detection of copy number abnormalities was obtained from the amplified or unamplified samples. The changes are also consistent with previous reports in the scientific literature (Macville et al., CANCER RESEARCH 59:141–150, 1999; Kloth et al., BMC GENOMICS 8:53, 2007).

A Reliable, Scalable Source of Faithfully Amplified Genomic DNA for Your Research Today and Tomorrow

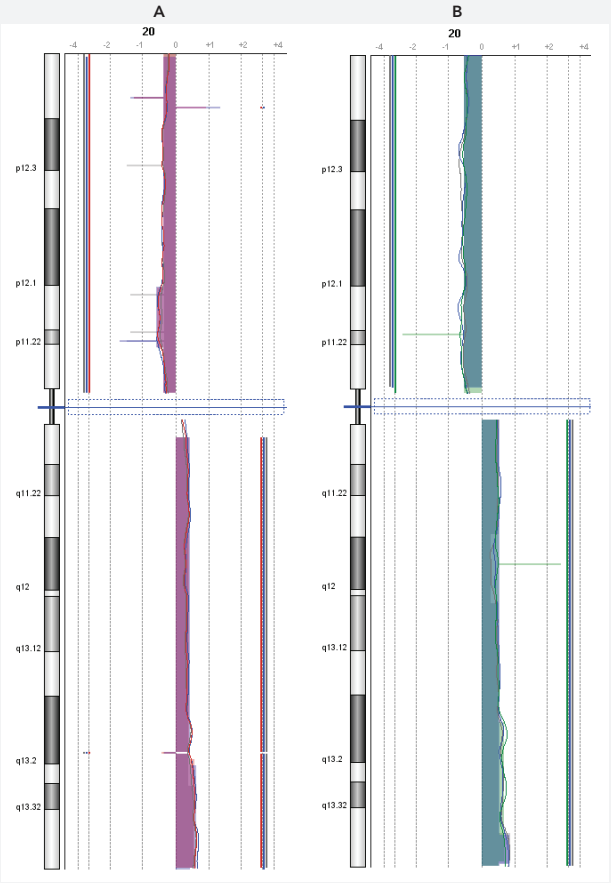
Genomic analysis platforms are evolving quickly. Ovation WGA System offers a linear amplification technology that enables the generation of a scalable resource that faithfully replicates valuable clinical samples, and is sustainable for your current and future research needs, independent of your genomic analysis platform.

In combination with the simplicity and robustness of the procedure, you can confidently and easily incorporate it in your research, regardless of the choice of analytical platform, selection of allele locus or patient sample. Ovation WGA System helps you expedite your discovery and research.

ORDERING INFORMATION

Ovation® WGA System	
Part No.	**No. of Reactions**
6100-12	12 Reactions
Technical Documents	
Ovation WGA System User Guide Ovation WGA System Quick Protocol	

FIGURE 5 **Chromosome Gains and Losses on CGH Arrays Consistent Between Amplified and Unamplified Samples**



Aberations (HeLa-Alexa 5, Male-Alexa 3) for chromosome 20. Agilent aCGH Analytics ideograms illustrating comparable results for unamplified genomic DNA (left, one µg) and Ovation WGA-amplified SPIA product (right, 10 ng).

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