GENOMIC DNA AMPLIFICATION

Genomic DNA Amplification Product Listing

REDTaq® Genomic DNA Polymerase

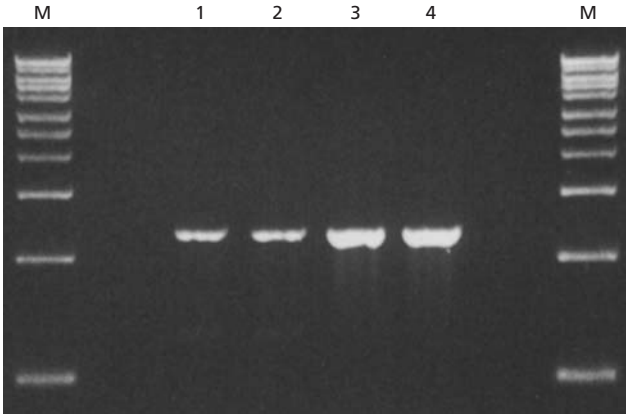
REDTaq Genomic DNA Polymerase is a special formulation of REDTaq DNA Polymerase designed to provide enhanced amplification of more complex genomic templates. REDTaq Genomic DNA Polymerase is more sensitive, produces higher yields and is more capable of generating longer product lengths. It has all the advantages of REDTaq DNA Polymerase, such as easy visualization of enzyme addition and complete reaction mixing, and direct loading to an agarose gel. The dye migrates slightly faster than bromophenol blue at the same rate as a 125 base pair fragment.

The inert red dye has no effect on automated or manual sequencing, restriction digestions, ligation, or other downstream applications. However, if dye removal is desired, this can easily be accomplished using any standard purification method.

Features and Benefits

- Enhanced amplification on genomic and difficult DNA templates
- No loading buffers or tracking dyes required. The PCR product is loaded directly onto an agarose gel after amplification
- Quick recognition when the REDTaq has been added to the reaction tube
- Confirms proper mixing at a glance for greater consistency across reactions
- PCR samples can be easily re-amplified as in nested PCR
- The unique red dye migrates slightly faster than bromphenol blue

Higher Yields from Genomic Templates with REDTaq Genomic DNA Polymerase



PCR reactions were set up using 1 µl of mouse genomic DNA and 1 unit of polymerase. The resulting amplicon is a specific 1181 bp fragment. Each sample was prepared in duplicate, and conditions for both sets were identical with the exception of the enzyme used.

Lanes M: 1 kb DNA Ladder (D3937)
Lanes 1, 2: REDTaq DNA Polymerase
Lanes 3, 4: REDTaq Genomic DNA Polymerase

Components: REDTaq Genomic DNA Polymerase
10× PCR Buffer or 10× PCR Buffer without $MgCl_2$
Separate vial of 25 mM $MgCl_2$ included with D2812

Unit definition: One unit incorporates 10 nmol of total dNTPs into acid-precipitable DNA in 30 min at 74 °C

Concentration: 1 unit per µl

Storage: –20 °C
Shipped in wet ice

Cat. No.	Product Description	Quantity
D8312	REDTaq Genomic DNA Polymerase with 10× Reaction Buffer containing $MgCl_2$	50 units 250 units 1,000 units 2,500 (10 x 250) units
D2812	REDTaq Genomic DNA Polymerase with 10× Reaction Buffer without $MgCl_2$. Includes a separate tube of 25 mM $MgCl_2$	50 units 250 units 1,000 units 2,500 (10 x 250) units

GENOMIC DNA AMPLIFICATION

The Universal Vectorette™ System *A PCR-based method for DNA walking and mapping*

The Vectorette system is a PCR-based method for DNA walking and mapping that uses a form of unidirectional PCR for amplifying and sequencing unknown genomic or large construct DNA. The system eliminates the time-consuming process of making and screening libraries to obtain overlapping clones and using conventional nucleic acid purification and screening procedures. A Vectorette unit is employed, which consists of a double stranded linker with an internal mismatched region and a sticky end.

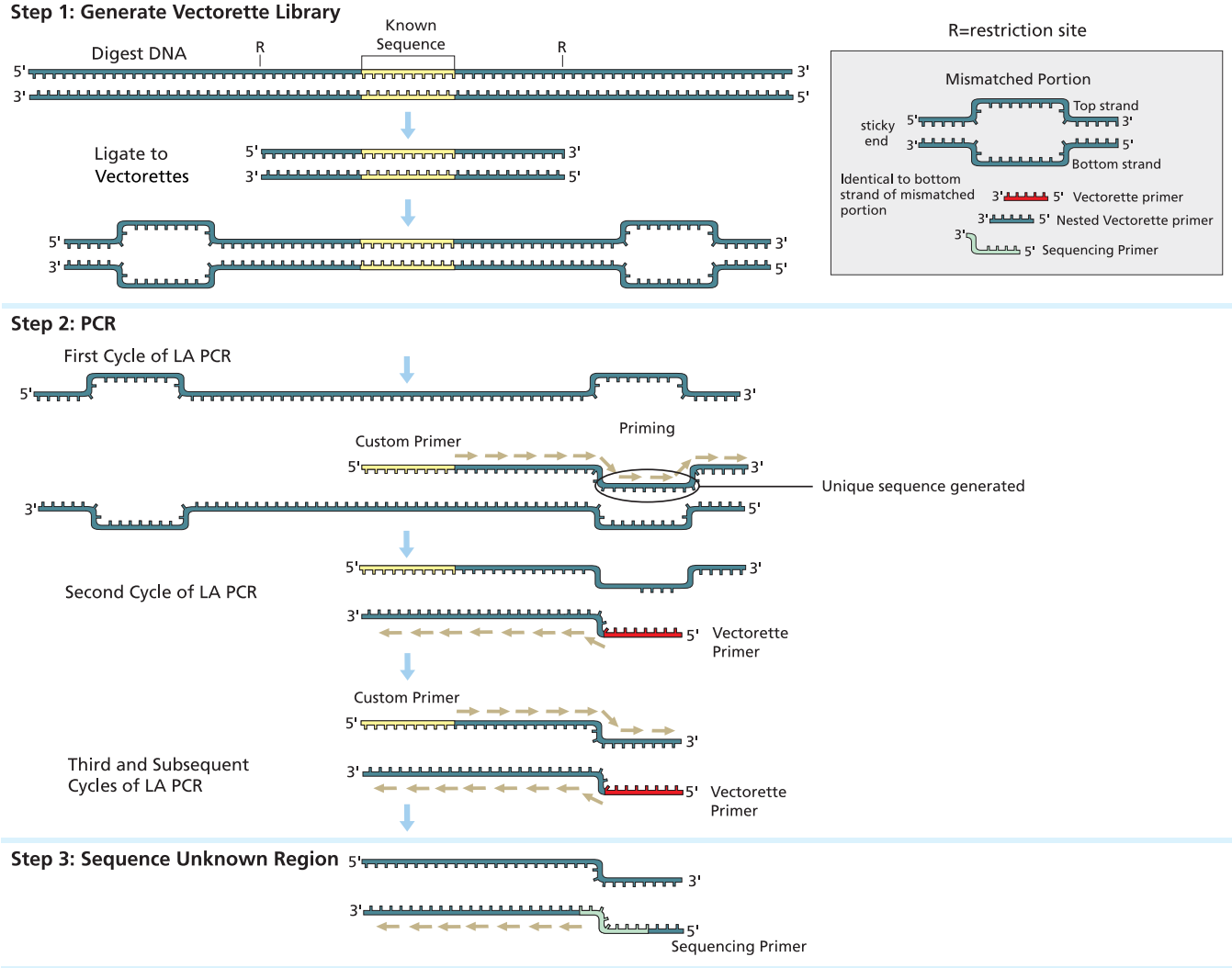
The Universal Vectorette system uses three simple steps to obtain DNA sequence information (Fig. 1):

Step 1: Genomic or large construct DNA containing the target sequence is digested with a restriction enzyme and Vectorette units are ligated to the 5′ and 3′ ends to create a Vectorette library.

Step 2: PCR is performed on the Vectorette library using a primer complementary to the mismatched region of the Vectorette unit (Vectorette primer provided) and a primer specific to the known DNA sequence. In the first PCR cycle, primer extension occurs only from the specific PCR primer that hybridizes to the known sequence in the DNA fragment within the Vectorette library. Extension from this primer generates a unique sequence as the polymerase reads through the mismatched portion of the Vectorette. Subsequent PCR cycles generate a DNA fragment between the known sequence and the Vectorette unit on the end of the fragment. Any Vectorette fragment that does not contain a sequence that is complementary to the specific primer will not generate a PCR product.

Step 3: A separate sequencing primer is included (slightly nested) that can be used to perform a sequencing reaction from the Vectorette end. PCR products are typically obtained from a single PCR run, however, nested primers are included to increase specificity when amplifying more complex templates. The PCR products generated by the Vectorette system can be used directly for cycle sequencing or cloned into commercially available vectors for further characterization.

Figure 1. Vectorette System Process



GENOMIC DNA AMPLIFICATION

The Universal Vectorette System offers the flexibility to generate Vectorette libraries from purified genomic DNA by *Bam*HI, *Cla*I, *Eco*RI, *Hind*III or blunt restriction enzyme digests. This system can be used with 1 µg template genomic DNA or less, and provides a time saving alternative to traditional library construction and screening. The protocol can also be modified for high throughput applications.

Ideal for:

Genome walking
Sequencing of yeast artificial chromosome (YAC) termini
Sequencing of cosmid insert termini
Mapping of promoters, introns, microsatellites, SSRs and STRs
Sequencing of large clones without sub-cloning
Mapping of regions containing deletions, insertions and translocations
Gap-filling in genome mapping projects
Identification of flanking genomic sequences of transgenes in transgenic organisms

Features and Benefits

- Cell-free gene manipulation replaces cloning and subcloning in many molecular genetics projects
- Two and three-step procedures can be performed in a single day
- High fidelity, highly specific amplifications up to 20 kb from genomic DNA
- Eliminates the need for nested PCR in most applications

Storage: –20 °C
Shipped in wet ice

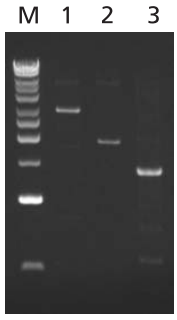


Figure 2. *Three different primers were used on a Cla I human genomic DNA Vectorette library to generate three different sizes of amplicons. The fragments generated are from different regions of the human globin gene.*

Lane M: 1 kb DNA Ladder (D3937)
Lane 1: 3 kb amplicon
Lane 2: 1.9 kb amplicon
Lane 3: 1.3 kb amplicon

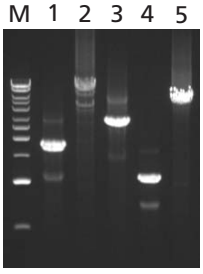


Figure 3. *Positive control PCR results for 5 different Vectorette libraries. This gel illustrates a common primer to a known sequence generating different amplicon size fragments on five different Vectorette libraries.*

Lane M: 1 kb DNA Ladder (D3937)
Lane 1: Bam HI Vectorette amplicon, 1.9 kb
Lane 2: Cla I Vectorette amplicon, 8.1 kb
Lane 3: Eco RI Vectorette amplicon, 3 kb
Lane 4: Hind III Vectorette amplicon, 1.1 kb
Lane 5: Sma I Vectorette amplicon, 4.8 kb

Cat. No.	Product Description	Quantity
UVS1	Universal Vectorette System 1 kit sufficient for 25 ligation reactions and 20 PCR reactions (50 µl reaction volume)	1 kit

GENOMIC DNA AMPLIFICATION

Human Genomic DNA

Human Random Control DNA Panels for use as reference standards

Sigma-Aldrich and ECACC have teamed together to provide researchers with control populations of human genomic DNA for gene regulation and quantitative PCR research. The range of Human Random Control (HRC) DNA samples represents a control population of 480 UK Caucasian blood donors. The HRC DNA is extracted from lymphoblastoid cell lines derived by Epstein Barr Virus (EBV) that can be continuously propagated in culture. This ensures an infinite supply of the unvarying DNA panels. The composition of each panel is completely defined and standardized so that each lot will be identical. Therefore, the HRC DNA Panels can be used as reference standards for routine quality control in the laboratory.

Features and Benefits

- Consistent Control Samples – The DNA is extracted from immortalized cell lines held as cryopreserved banks, so there is no batch-to-batch variation
- Convenient and Ready to Use – Ideal for most standard genetic research applications and avoids errors in preparing controls
- Cost Effective – The 10 µg format provides enough purified HRC DNA for thousands of PCR assays

The purified HRC DNA is available in 5 different panels (HRC1 through 5) consisting of 96 individuals and containing 10 µg DNA each at a concentration of 200 ng/µl. For convenience, three of the panels (HRC1, HRC2, and HGDBC1) are available in a PCR compatible 96-well format at 8 ng/µl, circumventing the need for aliquoting or dilution.

Human Genetic Disease DNA Panel for Breast Cancer

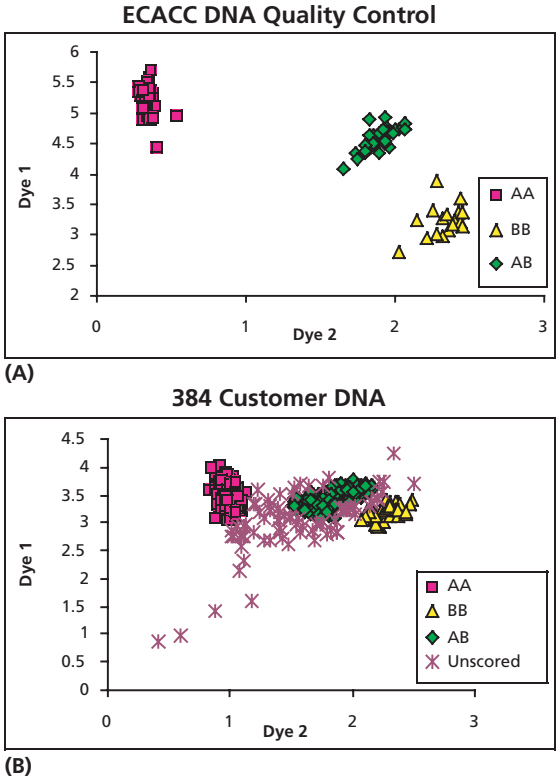
Also available is a Human Genetic Disease DNA Panel for Breast Cancer (HGDBC1). The samples are taken from female patients that have all been diagnosed as having breast cancer. The HGDBC1 panel is provided in a 96-well plate and can be used directly in automated gene analysis systems.

ecacc SIGMA

Working in Partnership

ecacc.org.uk **sigma-aldrich.com**

Increased effectiveness and reliability of HRC DNA Panels



Demonstration of how DNA quality can affect assay performance. The results of the 5' nuclease assay using 96 ECACC Human Random Control (HRC) DNA samples (plot A) and 384 Customer DNA samples (plot B). Homozygotes for allele 1 (AA, top left), heterozygotes for alleles 1 and 2 (AB, middle), homozygotes for allele 2 (BB, lower right). In plot A, using the ECACC HRC1 DNA, the lack of scatter and tight clustering of data points makes scoring the two alleles quite clear. Whereas in plot B, the wide scatter and lack of clustering, make scoring the different alleles very difficult, showing how DNA quality can affect assay performance. Data provided courtesy of MRC Geneservice, Cambridge.

Cat. No.	Product Description	Quantity
HRC1	Human Random Control Panel 1 Concentration: 200 ng/µl	10 µg
HRC2	Human Random Control Panel 2 Concentration: 200 ng/µl	10 µg
HRC3	Human Random Control Panel 3 Concentration: 200 ng/µl	10 µg
HRC4	Human Random Control Panel 4 Concentration: 200 ng/µl	10 µg
HRC5	Human Random Control Panel 5 Concentration: 200 ng/µl	10 µg
HRC1	Human Random Control Panel 1 Concentration: 8 ng/µl 96-well plate (50 µl/well)	1 each
HRC2	Human Random Control Panel 2 Concentration: 8 ng/µl 96-well plate (50 µl/well)	1 each
HGDBC1	Human Genetic Disease – Breast Cancer Concentration: 8 ng/µl 96-well plate (50 µl/well)	1 each

GENOMIC DNA AMPLIFICATION

Extract-N-Amp™ Plant PCR Kits

From leaf tissue to PCR in under 15 minutes

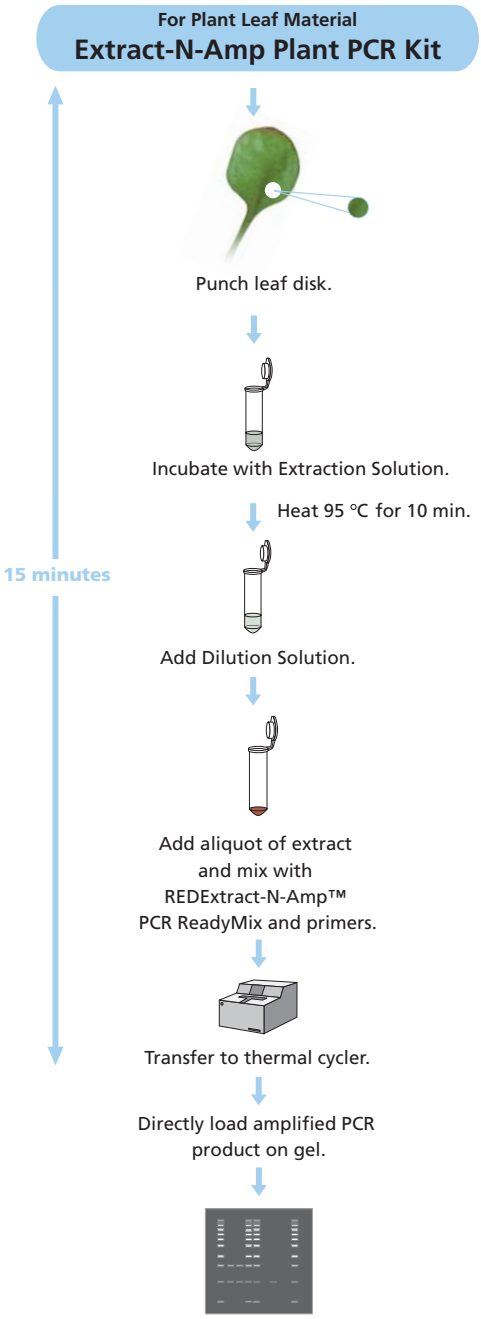
The Extract-N-Amp Plant PCR Kits contain all the reagents necessary to rapidly extract genomic DNA from plant leaves and amplify targets of interest by PCR. A novel extraction solution eliminates the need for conventional freezing of plant tissues with liquid nitrogen, mechanical disruption, organic extraction, column purification, or precipitation of DNA. The kit also includes a PCR reaction mix, specially formulated for amplification directly from extract. This formulation uses an antibody based hot start DNA polymerase for specific amplification. The PCR master mix comes in two formulations: Extract-N-Amp Plant PCR Mix and REDExtract-N-Amp Plant PCR Mix. The REDExtract-N-Amp PCR Mix contains a dye that acts as a tracking dye and allows for convenient direct loading of PCR reactions onto agarose gels for analysis.

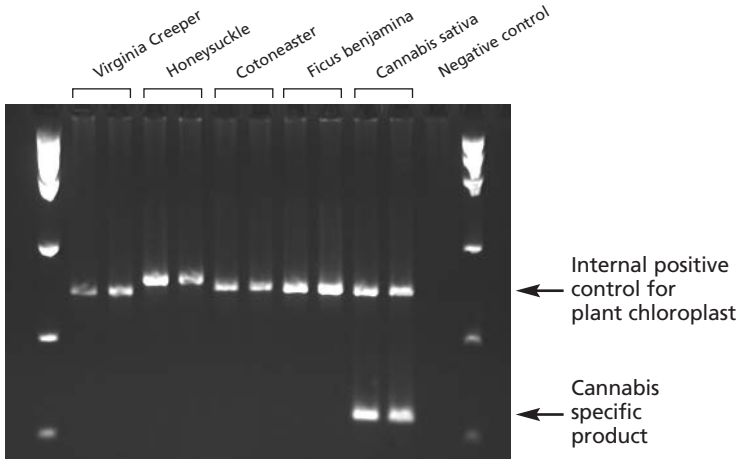
Genomic DNA is extracted from 0.5 to 0.7 cm plant leaf disks that have been cut with a standard paper punch and incubated in Extraction Solution at 95 °C for 10 minutes. An equal volume of Dilution Solution is added to the extract to neutralize inhibitory substances prior to PCR. A portion of the DNA extract is then added to a PCR reaction containing primers and either the REDExtract-N-Amp or Extract-N-Amp PCR Mix.

Features and Benefits

- Single-step extraction of plant genomic DNA for PCR in less than 15 minutes
- No freezing, mechanical disruption, organic extraction, column purification or precipitation required
- Specially formulated PCR master mix for use with plant extract
- Antibody inactivated DNA polymerase for highly specific PCR amplification of genomic DNA
- REDExtract-N-Amp requires no loading buffers or tracking dyes for gel analysis
- Compatible with high-throughput requirements for genetic analysis of plants
- Plant extract storage at 4 °C for up to 6 months (Fig. 3)

Storage: –20 °C
Shipped in wet ice
R: 36/37/38 S: 26-36





PCR analyses of genomic DNA extracted from 5 different plant species using Sigma's Extract-N-Amp Plant Kit

Figure 1. *Extract-N-Amp Plant PCR Kit used to isolate and amplify genomic DNA from various plant sources. Genomic DNA was extracted from 0.5 cm leaf disks that were cut using a standard paper punch. DNA was extracted using the Extract-N-Amp Plant PCR Kit in less than 15 minutes. All samples were then amplified using the specially formulated hot start PCR mix. The products were generated from a 30-cycle duplex reaction containing primers specific to plant chloroplast (upper band) and primers specific to Cannabis sativa DNA (lower band). MW ladder is 100, 200, 400 and 800 bp. Data provided by Andy Hopwood, Forensic Science Service, Birmingham, England.*

GENOMIC DNA AMPLIFICATION

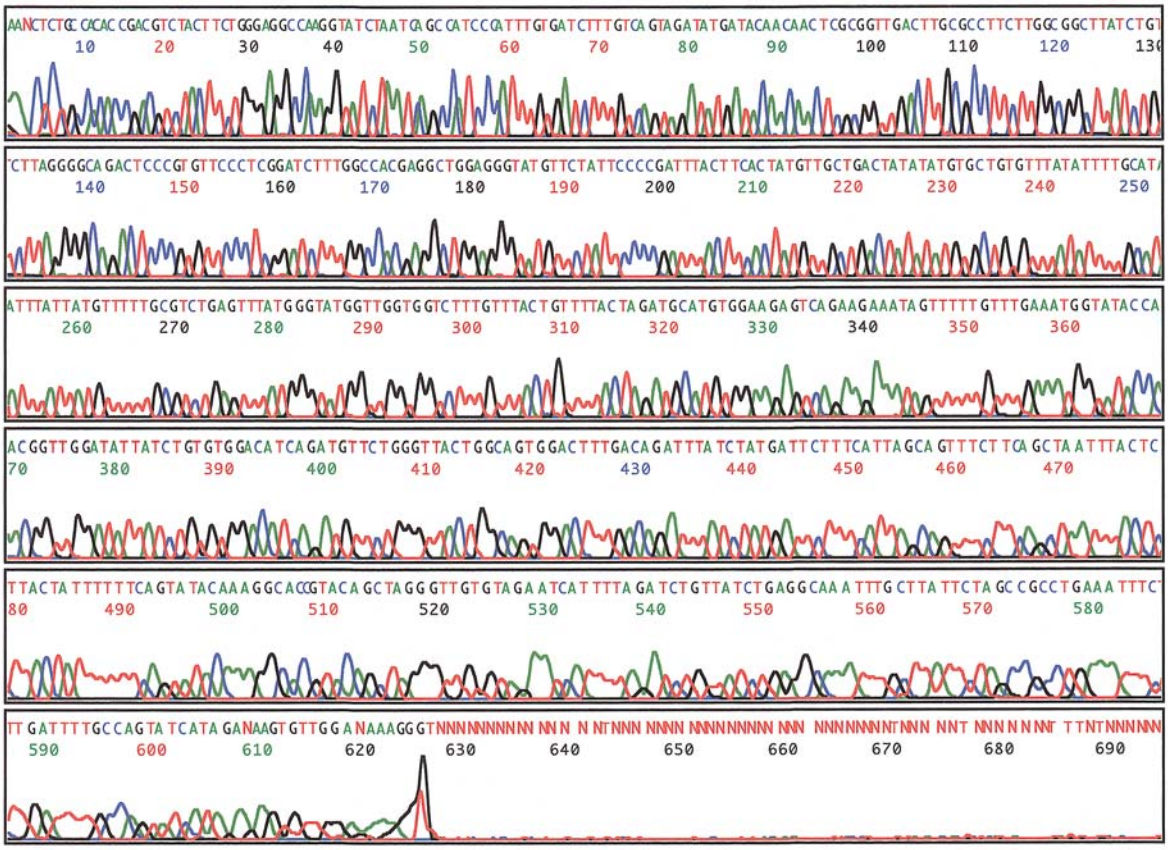
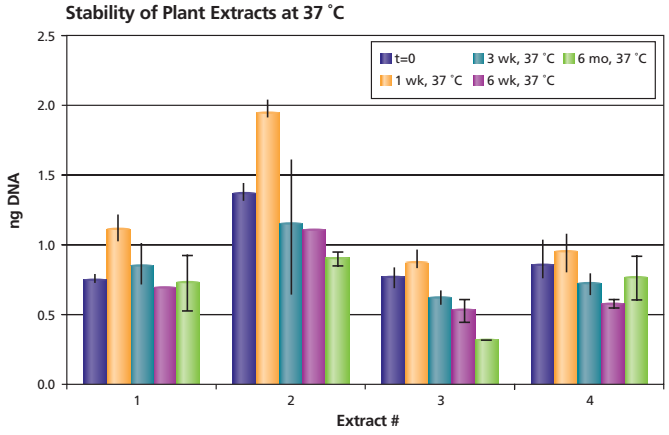


Figure 2. Sequence was resolved on a ABI 3100 from a purified, 645 bp corn leaf PCR product. The PCR product was purified with the GenElute PCR Clean-Up Kit. The DNA extraction and PCR were performed using Sigma's Extract-N-Amp Plant PCR kit. The sequence was obtained by using ABI BigDye® Terminator Chemistry and the same primers as for the original PCR.



Stability of DNA in corn leaf extracts

Figure 3. Eight disks were punched from a corn leaf, and DNA was extracted according to the procedure in the Technical Bulletin for the Extract-N-Amp Plant Kit. Two 4-µl aliquots from each were analyzed immediately by quantitative PCR with SYBR Green detection on an ABI Prism 7700. DNA standards for quantitative PCR were purified DNA prepared from corn leaf tissue with the GenElute Plant Genomic DNA kit (Catalog Number G2N70). Half of the leaf extracts were stored at 4 °C (recommended storage conditions) and the other half at 37 °C (accelerated storage). Quantitative PCR was repeated after 1, 3, and 6 months from extracts at 4 °C, and after 1 week, 3 weeks, 6 weeks, and 6 months from extracts at 37 °C. Results for storage at 37 °C are shown. The average of 2 replicate PCR assays from each extract is plotted. Error bars represent one standard deviation. Results for storage at 4 °C are essentially the same as those shown for 37 °C.

Cat. No.	Product Description	Extractions	Amplifications
XNAPS	REDExtract-N-Amp Plant PCR Kit	10	10
XNAP	REDExtract-N-Amp Plant PCR Kit	100	100
XNAPE	REDExtract-N-Amp Plant PCR Kit	100	500
XNAPR	REDExtract-N-Amp Plant PCR Kit	1000	1000
XNAPRE	REDExtract-N-Amp Plant PCR Kit	1000	5000
XNAP2	Extract-N-Amp Plant PCR Kit	100	100
XNAP2E	Extract-N-Amp Plant PCR Kit	100	500
XNAR	Extract-N-Amp Plant PCR Kit	1000	1000
XNAP2RE	Extract-N-Amp Plant PCR Kit	1000	5000

GENOMIC DNA AMPLIFICATION

Extract-N-Amp™ Blood PCR Kits

From whole blood to PCR in under 8 minutes

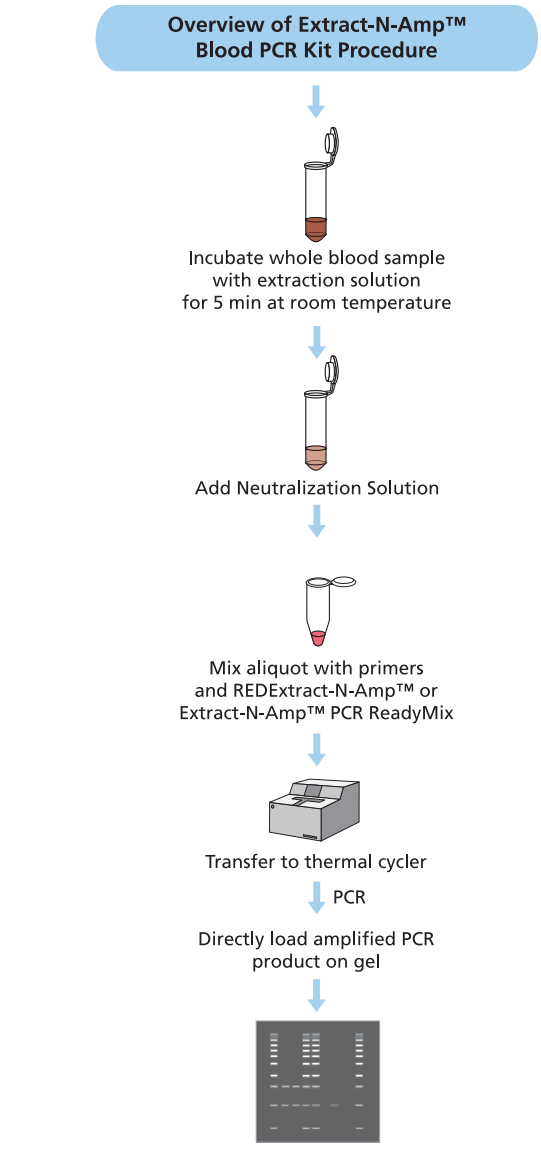
The Extract-N-Amp Blood PCR Kits contain all of the reagents necessary to rapidly extract genomic DNA from whole blood and amplify targets of interest by PCR. This novel extraction system eliminates the need for any type of purification, organic extraction, centrifugation, heating, filtration or alcohol precipitation. The kit also includes a PCR master mix, especially formulated for amplification directly from the extract. This formulation uses an antibody based hot start DNA polymerase for specific amplification. The PCR master mix comes in two formulations: Extract-N-Amp Blood PCR Mix and REDExtract-N-Amp Blood PCR Mix. The REDExtract-N-Amp Blood PCR Mix contains a tracking dye that allows for convenient direct loading of PCR reactions onto agarose gels for analysis.

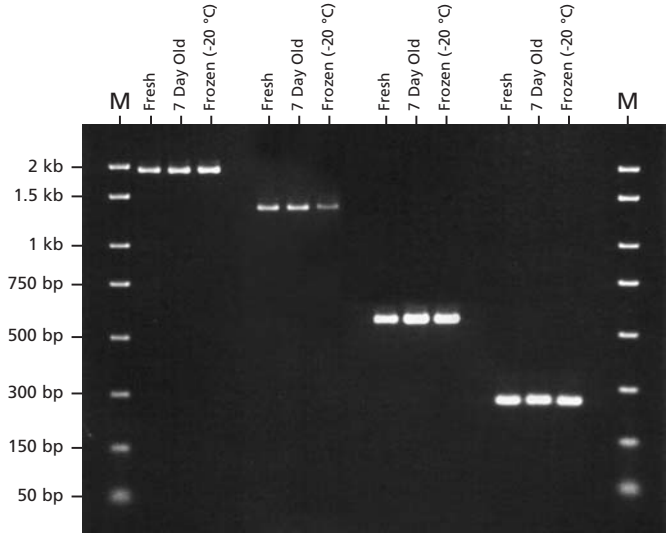
Genomic DNA is extracted from 10 µl of whole blood by simply adding the Extraction Solution and incubating for 5 minutes at room temperature. The Neutralization Solution is added to the extract to counteract inhibitory substances prior to PCR. A portion of the DNA extract is then added to the specially formulated PCR mix.

Features and Benefits

- Efficient 8 minute prep allows greater speed and throughput
- No need for any type of purification, organic extraction, centrifugation or alcohol precipitation
- Simple, 3 step procedure with no special equipment required
- Antibody inactivated DNA polymerase included for highly specific PCR amplification of genomic DNA
- Compatible with any format (single tube, 96-well, etc.)
- No phenol/chloroform extraction required
- Blood extract storage at 4 °C for up to 6 months (Fig. 2)

Storage: –20 °C
Shipped in wet ice
R: 34 S: 26-27-36/37/39





PCR analysis of genomic DNA isolated from blood using Sigma's Extract-N-Amp Blood PCR Kit

Figure 1. *Extract-N-Amp Blood PCR Kit used to isolate genomic DNA from fresh, 7 day old, & frozen blood. DNA was extracted and neutralized from 10 µl of whole blood in 5 minutes at room temperature using the REDExtract-N-Amp Blood PCR kit. The PCR products were then generated using the specially formulated hot start PCR mix included in the kit. PCR products generated are 1.8 kb for carnitine palmitoyltransferase II, 1.3 kb for a mitochondrial DNA control region, 547 bp for human surfactant protein B, and 320 bp for the 5' untranslated region of human major histocompatibility complex class II.*

GENOMIC DNA AMPLIFICATION

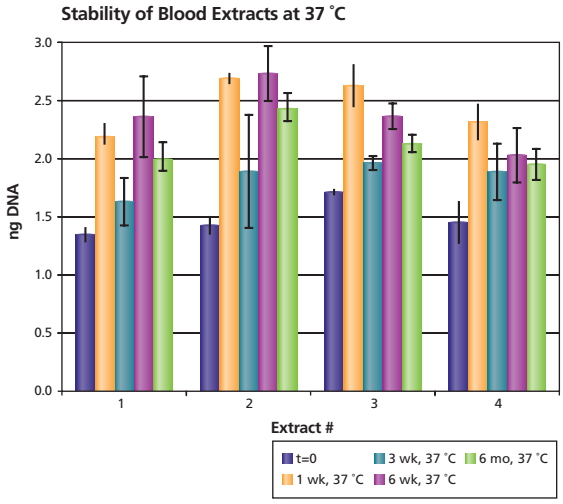


Figure 2. *Stability of Extract-N-Amp Blood Extracts. Blood was drawn from 2 human volunteers into Vacutainer® tubes containing EDTA. Extractions were performed in duplicate providing 4 samples total. Samples were stored at 37 °C and removed at various time intervals for testing. Stability was determined by monitoring yield from quantitative PCR using an ABI 7700 instrument. The DNA standards used for the quantitative PCR were generated from the same blood draw as the test samples. DNA for the standards was purified using the GenElute Blood Genomic DNA Kit (NA2000) and stored as single aliquots at –20 °C. The PCR products were generated using primers for a 547 bp product for human surfactant protein B (SPB; Lin & Floros, 2000, BioTechniques, 29: 460-466). The results clearly show no loss of amplification of the SPB PCR product even after storage at 37 °C for 6 months.*

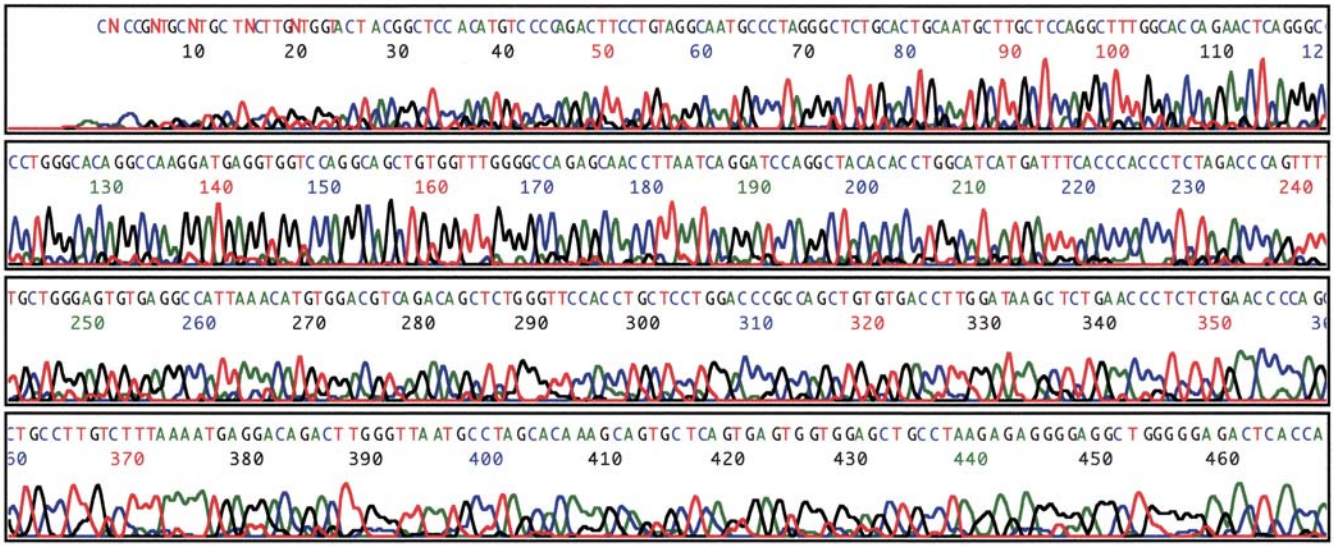


Figure 3. *Direct sequence from PCR products generated using the Extract-N-Amp Blood Kit. A 547 bp product for human surfactant protein B was generated using the Extract-N-Amp Blood PCR Kit. The product was sequenced directly using BigDye terminator chemistry. Sequencing reactions were resolved on an ABI 3100.*

Note: Some PCR products require further clean-up prior to sequencing. The GenElute PCR Clean-Up Kit (NA1020) is recommended.

Cat. No.	Product Description	Extractions	Amplifications
XNABS	REDExtract-N-Amp Blood PCR Kit	10	10
XNAB	REDExtract-N-Amp Blood PCR Kit	100	100
XNABE	REDExtract-N-Amp Blood PCR Kit	100	500
XNABR	REDExtract-N-Amp Blood PCR Kit	1000	1000
XNABRE	REDExtract-N-Amp Blood PCR Kit	1000	5000
XNAB2	Extract-N-Amp Blood PCR Kit	100	100
XNAB2E	Extract-N-Amp Blood PCR Kit	100	500
XNAB2R	Extract-N-Amp Blood PCR Kit	1000	1000
XNAB2RE	Extract-N-Amp Blood PCR Kit	1000	5000

GENOMIC DNA AMPLIFICATION

Extract-N-Amp™ Tissue PCR Kits

From tissue or cells to PCR in 15 minutes

The Extract-N-Amp™ Tissue PCR Kits provide all the reagents necessary to rapidly extract DNA from a wide variety of cells and tissues and amplify targets of interest by PCR (Fig. 1). A novel extraction method eliminates the need for long enzymatic digestions or homogenization. The kit also includes a specially formulated hot start PCR ReadyMix™ for amplification directly from the extract. The PCR ReadyMix comes in two formulations: Extract-N-Amp PCR ReadyMix and REDExtract-N-Amp PCR ReadyMix. The REDExtract-N-Amp PCR ReadyMix contains an inert dye that acts as a tracking dye and allows for convenient loading of PCR reactions onto agarose gels for analysis.

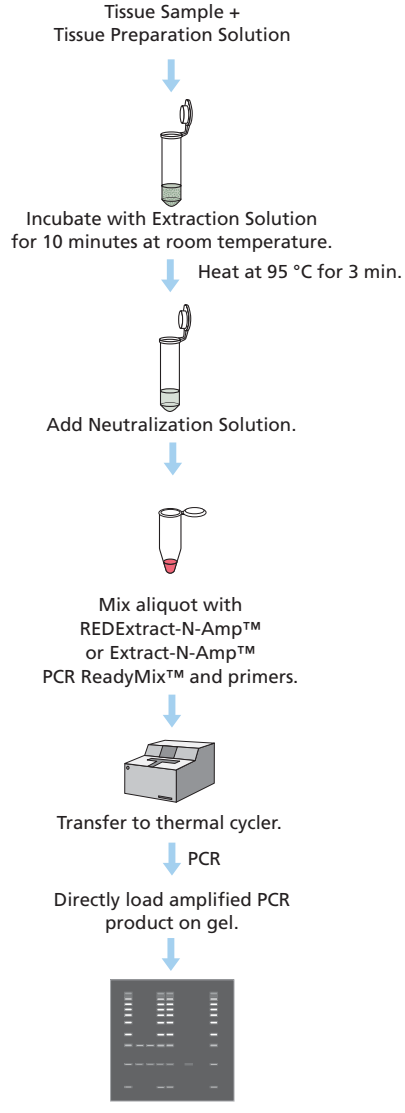
The kit comes with validated protocols to extract and amplify genomic DNA from mouse-tails, hair, animal tissue, saliva, and buccal swabs. In a typical procedure, genomic DNA is extracted from a tissue sample that has been incubated in the tissue preparation solution and extraction solution for 10 minutes at room temperature. The sample is heated to 95 °C for 3 minutes and then mixed with a third solution to neutralize inhibitory substances prior to PCR. A portion of the DNA extract is then added to a PCR reaction containing primers and either the REDExtract-N-Amp or Extract-N-Amp PCR ReadyMix included in the kit.

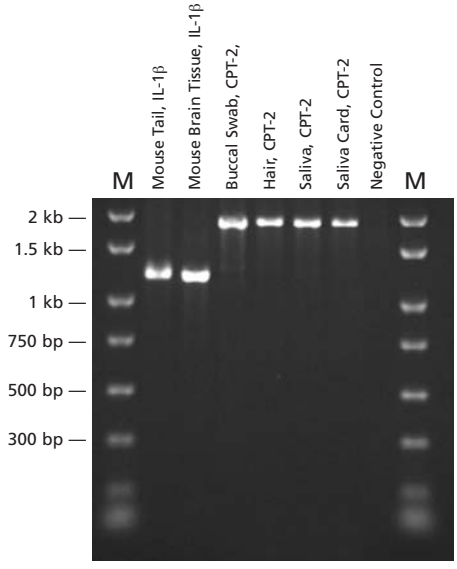
Feature and Benefits

- Fast – rapid extraction of genomic DNA for PCR in 15 minutes
- Convenient – no long enzymatic digestions
- Practical – perfect for quick genomic DNA isolation for genotyping
- Flexible – protocols available for mouse-tails, hair, animal tissue, saliva, and buccal swabs
- Specific – hot start antibody for highly specific PCR amplification of genomic DNA

Storage: –20 °C
Shipped in wet ice
R: 36/37/38-42/43 S: 26-36

Overview of Extract-N-Amp™ Tissue PCR Kit Procedure





PCR analysis of genomic DNA extracted from various samples using Sigma's Extract-N-Amp™ Tissue PCR Kit

Figure 1. *The Extract-N-Amp™ Tissue PCR Kit was used to extract and amplify genomic DNA from various sources. Genomic DNA was extracted from the samples using the appropriate protocol for each sample type as described in the Extract-N-Amp Tissue Technical Bulletin. In all cases, the extraction procedure was completed in less then 15 minutes. The extracted DNA was then amplified using the specially formulated hot start PCR ReadyMix™. The products generated are 1181 bp for the Interleukin 1 Beta gene in mouse and 1820 bp for the Carnitine palmitoyltransferase II in human. Markers are PCR Marker (Catalog Number P9577).*

GENOMIC DNA AMPLIFICATION

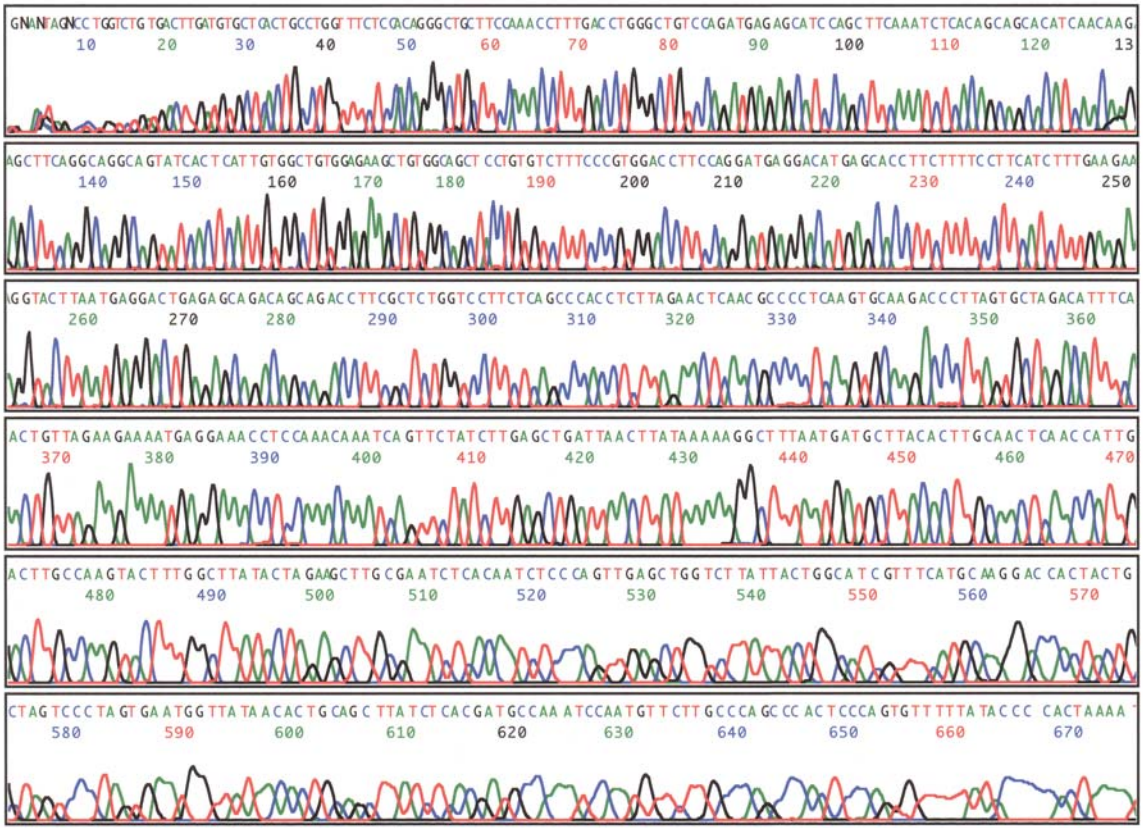
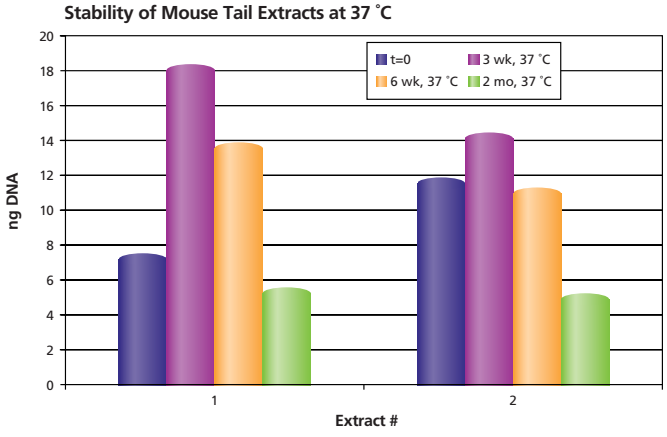


Figure 2. Sequence determination for 1181 bp Interleukin 1 Beta mouse-tail PCR product. *The PCR product was purified with the GenElute™ PCR Clean-Up Kit (Catalog Number NA1020). The DNA extraction and PCR were performed using Sigma's Extract-N-Amp™ Tissue PCR Kit. The sequence was obtained using the ABI BigDye® Terminator Chemistry and the same primers as for the original PCR. Reaction products were resolved on an ABI 310.*



Stability of DNA in mouse-tail extracts

Figure 3. *Mouse-tail samples were extracted according to the procedure in the Technical Bulletin for the Extract-N-Amp™ Tissue PCR Kit. The remaining mouse-tail tissue was removed from the samples for storage. 4 µl aliquots were analyzed immediately by quantitative PCR with SYBR® Green detection on an ABI Prism® 7700. DNA standards for quantitative PCR were purified DNA prepared from mouse tails using the GenElute Mammalian Genomic DNA kit (Catalog Number G1N70) and stored as single use aliquots at –20 °C. The mouse-tail extracts were stored at 37 °C (accelerated storage). Quantitative PCR was repeated after 3 weeks, 5 weeks and 2 months from extracts at 37 °C. Results for storage at 37 °C are shown. These results suggest that extracts will be stable for at least 6 months at the recommended storage temperature of 4 °C.*

Cat. No.	Product Description	Extractions	Amplifications
XNATS	REDExtract-N-Amp Tissue PCR Kit	10	10
XNAT	REDExtract-N-Amp Tissue PCR Kit	100	100
XNATR	REDExtract-N-Amp Tissue PCR Kit	1000	1000
XNAT2	Extract-N-Amp Tissue PCR Kit	100	100
XNAT2R	Extract-N-Amp Tissue PCR Kit	1000	1000

Inquire for bulk and high-throughput needs.

GENOMIC DNA AMPLIFICATION

SYBR® Green Extract-N-Amp™ Tissue PCR Kit

The SYBR Green Extract-N-Amp Tissue PCR Kit contains all the reagents needed for rapid extraction, amplification and detection of genomic DNA from mouse-tails and other animal tissues, buccal swabs, hair shafts, and saliva.

The SYBR Green Extract-N-Amp Tissue PCR Kit offers an innovative extraction system that eliminates the need for either long enzymatic digestions or homogenization. The product includes a specially formulated Hot Start SYBR Green PCR ReadyMix™ for amplification and quantitation directly from the extract.

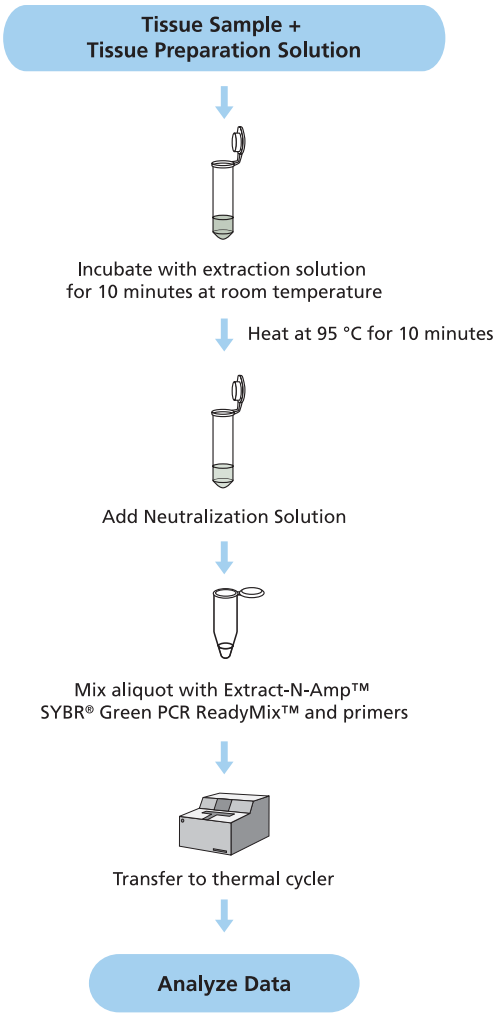
Procedure: DNA is rapidly extracted from a tissue by incubating the sample with a mixture of the Extraction Solution and the Tissue Preparation Solution at room temperature for 10 minutes. After a 3-minute heat denaturing step, an equal volume of Neutralization Solution B is added to the extract to neutralize inhibitory substances. The extract is ready for real-time PCR in any plate-based real-time thermal cycling system in less than 15 minutes!

Application: Ideal for genotyping, gene copy number experiments, and amplifying and quantifying DNA from multiple tissue sample types.

Features and Benefits

- Novel – all liquid, single-step extraction of genomic DNA for quantitative PCR (QPCR)
- Fast – tissue to QPCR in 15 minutes
- Convenient – no long enzymatic digestions and no column purifications
- Simple – rapid, easy-to-follow protocol
- Sensitive – specially formulated Hot Start SYBR Green PCR ReadyMix™ for highly specific PCR amplification and quantitation of genomic DNA
- Safe – no organic extraction with hazardous chemicals

Storage: –20 °C
R: 42 S: 36/37-45



Distinguish Differences in Gene Copy Number

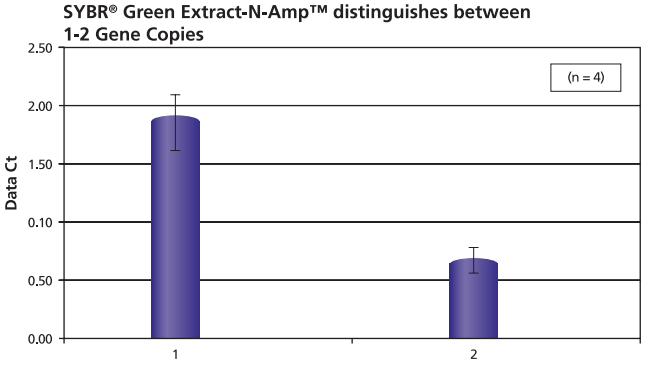


Figure 1. SYBR Green Extract-N-Amp Used to Distinguish between 1-2 Gene Copies. *Extracts were prepared following the standard protocol for the SYBR Green Extract-N-Amp Tissue PCR Kit from 8 mouse tails that contained either a single or double copy of the Diap2 gene. Two single-gene PCR reactions were run on each extract using the 2x Extract-N-Amp SYBR Green PCR ReadyMix.*

GENOMIC DNA AMPLIFICATION

Flexible Enough for A Wide Variety of Tissue Types

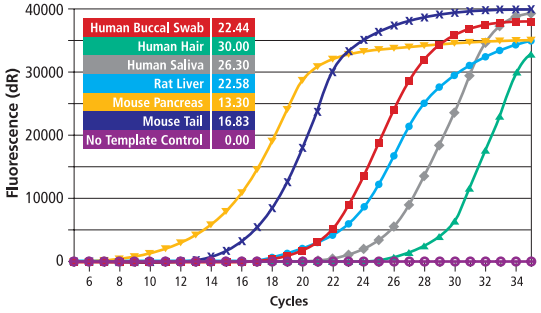


Figure 2. Quantitative PCR of DNA isolated from a variety of tissue types and sources. *The Extract-N-Amp Tissue PCR Kit was used to extract and amplify genomic DNA from various sources. The extracted DNA was then amplified using the specially formulated Hot Start SYBR Green PCR ReadyMix.*

High Sensitive SYBR Green PCR ReadyMix

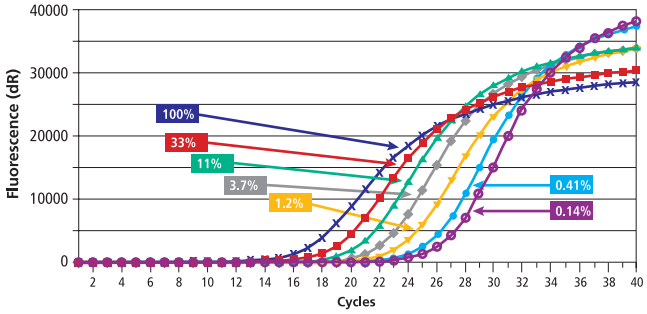


Figure 3. Quantitative PCR of DNA isolated from a series of decreasing extract dilutions. *DNA was extracted from a mouse-tail snip following the standard protocol for the SYBR Green Extract-N-Amp Tissue PCR Kit. The extract was diluted in decreasing three-fold increments, from 100% extract to 0.14% extract. As depicted above, message can be clearly detected even with the most dilute extract.*

Cat. No.	Product Description	Size	Quantity
XNATSG	SYBR Green Extract-N-Amp Tissue PCR Kit	10	1 kit
XNATG	SYBR Green Extract-N-Amp Tissue PCR Kit	100	1 kit
XNATRG	SYBR Green Extract-N-Amp Tissue PCR Kit	1,000	1 kit

GENOMIC DNA AMPLIFICATION

Extract-N-Amp™ Seed PCR Kits

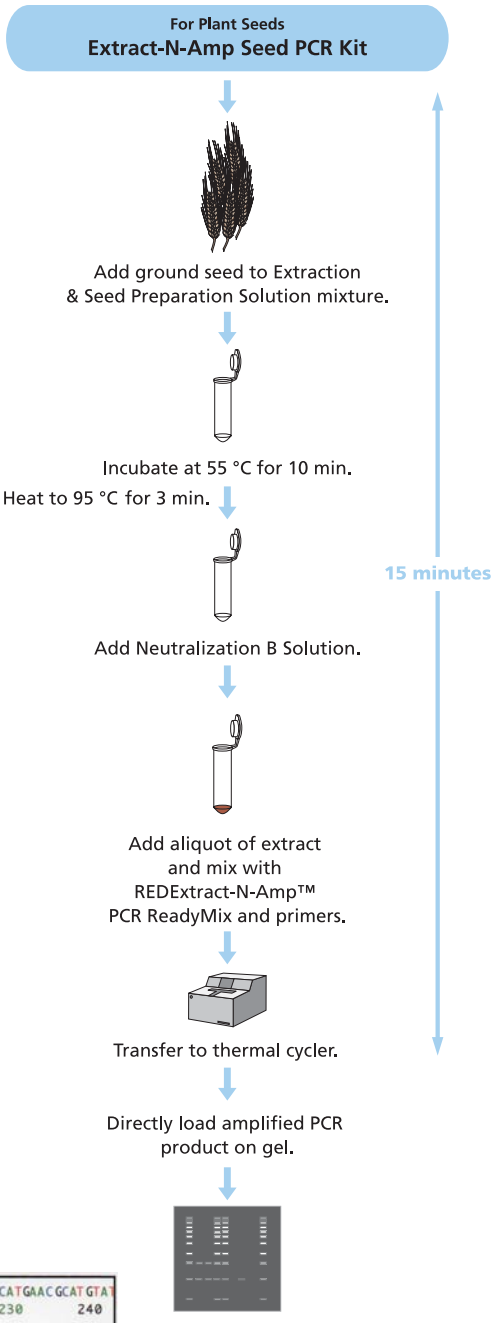
Rapid genomic DNA extraction from seeds

The Extract-N-Amp Seed PCR Kits can be used to extract seed genomic DNA that is suitable for PCR in just 15 minutes, using a straightforward extraction protocol. The PCR product that is amplified using the Extract-N-Amp Seed PCR Kit is suitable for direct sequencing. No phenol/chloroform extraction or alcohol precipitation is required. The DNA extracted by the Extract-N-Amp Seed PCR Kit is stable for at least 6 months at 4 °C, allowing for multiple reassays.

Features and Benefits

- Fast – 15 minute extraction of genomic DNA for PCR
- Flexible – compatible with a wide variety of seed sources
- Specific – Hot Start antibody for highly specific PCR amplification of genomic DNA
- Simple – no column purification or centrifugation required

Storage: –20 °C
R: 36/37/38-42 S: 26-36



Identify Specific Genes Before Planting Seeds

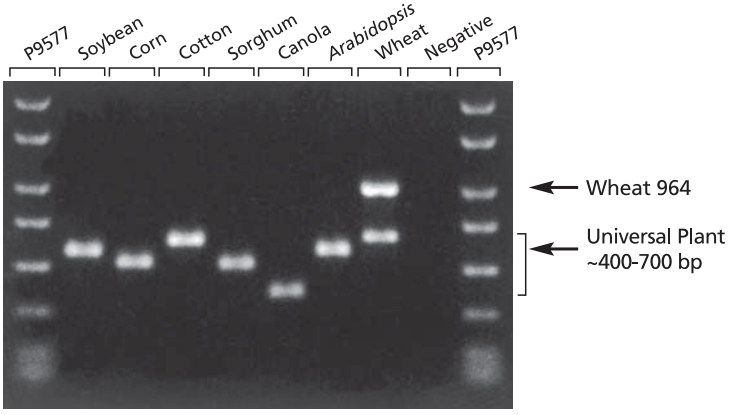


Figure 1. Genomic DNA was extracted from seeds using the protocol as described in the Extract-N-Amp Seed Technical Bulletin. *All extracts were then amplified using the specially formulated JumpStart™ PCR ReadyMix and PCR primers multiplexed for both a universal chloroplast gene (~400-700 bp) and the acetylcoenzyme A carboxylase gene specific to wheat (964 bp).*

Sequence PCR Products for Further Characterization of Genes

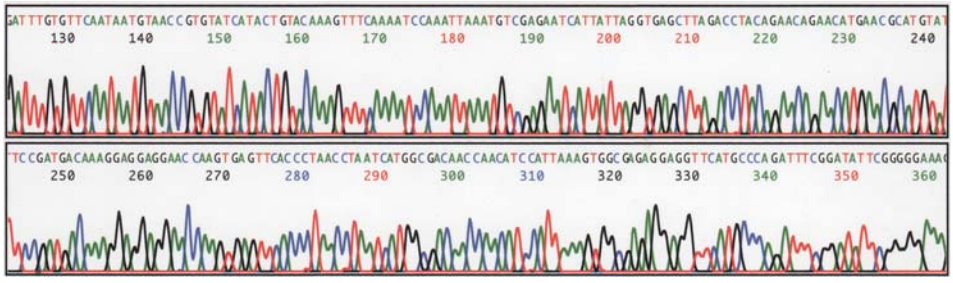


Figure 2. DNA extraction from wheat and subsequent PCR were performed using Sigma's Extract-N-Amp Seed PCR Kit (see Fig. 1 legend). *The PCR product of interest was purified with the GenElute PCR Clean-Up Kit (Catalog Number NA1020). The chromatogram shows a portion of the sequence determination for the 964 bp acetyl-coenzyme A carboxylase wheat PCR product. The sequence was obtained using the ABI BigDye® Terminator Chemistry and the same primers as for the original PCR. Reaction products were resolved on an ABI 377.*

Cat. No.	Product Description	Extractions	Amplifications
XNASS	REDExtract-N-Amp Seed PCR Kit	10	10
XNAS	REDExtract-N-Amp Seed PCR Kit	100	100
XNAS2	Extract-N-Amp Seed PCR Kit	100	100