



A Maximum Instrument at a Minimum Size

Introducing the Maxwell™ 16 Instrument: A Simple, Robust and Flexible Tool for DNA Purification

By Dan Kephart, Ph.D., Steve Krueger, B.S.ChE, Terri Grunst, M.B.A., and Hemanth Sheno, Ph.D., Promega Corporation

Abstract

The Maxwell™ 16 Purification System combines compact instrumentation, prefilled Reagent Cartridges and optimized automated methods to maximize performance and flexibility while minimizing hands-on time required for DNA purification. The instrument is designed to process up to 16 samples in a single run of about 30 minutes. The purified DNA is ready for downstream analysis without sample preprocessing, centrifugation, precipitation or rehydration of a DNA pellet. In this article, we discuss the development of the Maxwell™ 16 System and demonstrate its use for purification of genomic DNA.

The Maxwell™ 16 Instrument is a small and extremely easy-to-operate system. The unique Reagent Cartridge facilitates genomic DNA purification from a wide variety of sample types.

Introduction

Genomic DNA (gDNA) purification is a routine procedure in the modern molecular biology laboratory. Purified DNA is commonly used in genotyping, mapping or gene structure analysis. Currently, scientists use many different methods for gDNA purification across a range of sample types, sizes and throughputs. High-throughput users have increasingly turned to automation to improve productivity. Low- to moderate-throughput users can spend as much as 25–30% of their time on DNA purification, yet have traditionally had few if any options for automation that meet their throughput needs without requirements of considerable capital investment, training, upkeep and maintenance. In large laboratories there is a need for less expensive “benchtop” equipment that can be dedicated to certain tasks, certain areas of a laboratory or even sites separated geographically from the main laboratory to accommodate workflow requirements.

To accomplish the goals of eliminating hands-on time and labor while providing high yield and purity of DNA, Promega developed the easy-to-use Maxwell™ 16 System. This system, composed of the Maxwell™ 16 Instrument and Kits containing prefilled Reagent Cartridges, was developed for the low- to moderate-throughput user. The Maxwell™ 16 Instrument is a small and extremely easy-to-operate system. The unique

Reagent Cartridge facilitates DNA purification from a wide variety of sample types. Future reagent kits will provide a broad range of purification applications to allow adaptation of the Maxwell™ 16 System to your changing needs.

Instrument Design Improves Productivity

The Maxwell™ 16 Instrument has a compact design (measuring 31.4 × 37.4 × 29.5cm) that minimizes the amount of valuable laboratory bench space it occupies (Figure 1, Panel A). Optimized methods are preloaded onto the instrument; once unpacked and plugged in, it is ready for use in DNA purification.

The functionality of the Maxwell™ 16 Instrument is based on the sequential capture and release of MagneSil® Paramagnetic Particles (PMPs) into the wells of the Reagent Cartridges, which are supplied loaded with predispensed reagents (Figure 1). The instrument uses a powerful magnet and unique Plunger design to lyse and capture target material (i.e., genomic DNA) while washing away impurities. The ability of the instrument to selectively capture and move the PMPs during processing eliminates the need for complex liquid handling and reagent bottles that can clog or become contaminated. The Reagent Cartridge and Plunger come into contact with a single sample; they are removed and discarded after each run. Single-use Reagent Cartridges also ensure that new reagents are delivered untainted to maximize performance and run reproducibility.

Generally 1–16 samples can be processed in about 30 minutes using the Maxwell™ 16 Instrument. The unique Plunger design allows most sample types, including animal and plant tissue, to be purified without the need for preprocessing, such as proteinase K or other enzyme treatment, mechanical grinding, fractionation of white cells or centrifugation. For example, up to 1.2cm of mouse tail or 25–50mg of tissue are routinely placed directly into the cartridge for DNA purification. The ability to skip preprocessing saves hours compared to the normal routine of grinding or proteinase K digestion required for other sample preparation methods. After adding the sample, the Reagent Cartridges are placed into the Maxwell™ 16 Instrument, and simple on-screen prompts guide you through selection of the appropriate optimized method. When the Maxwell™ 16 Instrument run is completed, the purified DNA is ready for downstream analysis.

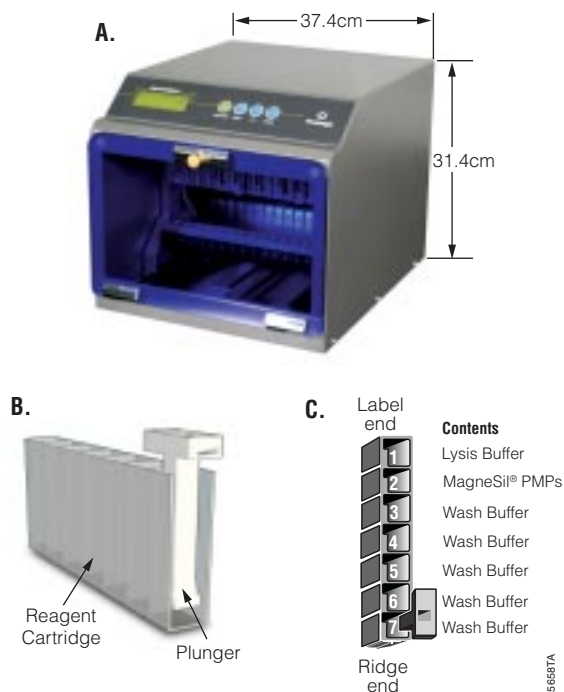


Figure 1. The Maxwell™ 16 Instrument is used with optimized reagents predispensed into disposable cartridges. Panel A. The Maxwell™ 16 Instrument. **Panel B.** The Maxwell™ 16 Reagent Cartridge, cut away to show the Plunger. The unique design of the cartridge allows direct processing of a variety of liquid and solid sample types with no need for preprocessing. **Panel C.** Top view of Maxwell™ 16 Cartridge.

Purified DNA is Ready for Downstream Applications

Blood is a relatively easy and often-used sample source for gDNA isolation. The Maxwell™ 16 Reagent Cartridges provided with the Maxwell™ 16 Blood DNA Purification Kit (Cat.# AS1010) are optimized to process a wide range of volumes of fresh or stored human blood as well as animal blood samples. Up to 400µl of whole blood can be processed (Figure 2) to yield approximately 8–16µg of gDNA from human blood, depending on the white blood cell (WBC) count. The purity of the gDNA is excellent, and its performance in downstream applications such as PCR is highly consistent (Figure 2). Starting volumes of blood can be reduced to as little as 10µl while retaining linear DNA recovery and excellent PCR results. DNA has also been purified from blood collected in citrate-, EDTA- or heparin-containing tubes with similar yield and purity results and no observed difference in application performance. Analysis in multiplexed PCR^(a) applications requiring highly pure DNA demonstrates the high quality of the Maxwell™ 16 Instrument-purified DNA.

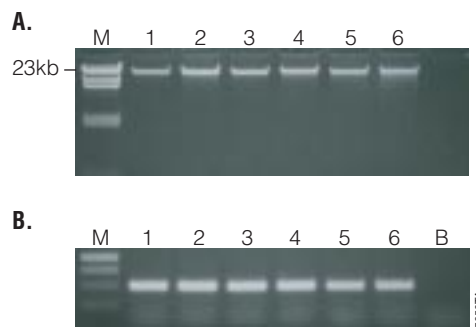


Figure 2. Isolation and analysis of DNA from human whole blood. DNA was isolated from 400µl blood samples obtained from 6 donors. **Panel A.** Aliquots (5µl) from each sample were resolved by agarose gel electrophoresis. Lane M, Lambda DNA/*Hind* III Markers (Cat.# G1711) **Panel B.** Equal volumes (1µl) of each sample were amplified using a primer pair specific for the human Factor II gene. Lane M, 1kb DNA Ladder (Cat.# G5711); Lane B, no-template PCR control. Average $A_{260/280}$ for samples was 1.72. Total eluted sample volume was approximately 210µl.

DNA isolation directly from whole blood using the Maxwell™ 16 Blood DNA Purification Kit minimizes the negative effects of storage conditions often encountered using other DNA purification systems. Normally, preprocessing requires collection of WBCs prior to DNA isolation, which increases hands-on time and can lead to significant loss of DNA from samples where cells are compromised due to poor storage or multiple freeze-thaw cycles. The Maxwell™ 16 System purifies DNA from blood stored at 4°C, -20°C or at room temperature; the system effectively captures DNA of a wide size distribution to maximize yield even from compromised samples.

Prefilled Reagent Cartridges for Different Sample Types

The Maxwell™ 16 System uses three different kits containing prefilled reagent cartridges to isolate gDNA from a wide variety of sample types including human and animal whole blood, white blood cell buffy coat, animal and plant tissue, eukaryotic and prokaryotic cells as well as whole organisms such as *Drosophila*. Animal blood and a variety of animal tissues have been used in the Maxwell™ 16 Instrument without the need for homogenization or prior processing. Whole tissue samples (up to 50mg) can be placed directly into the Maxwell™ 16 Reagent Cartridges supplied with the Maxwell™ 16 Tissue DNA Purification Kit (Figure 3). The elimination of preprocessing steps allows you to go from sample collection to data analysis in one day instead of two, greatly improving your productivity. DNA purified from a variety of tissues performs well in downstream applications (Figure 3).

Maxwell™ 16 for DNA Purification... continued

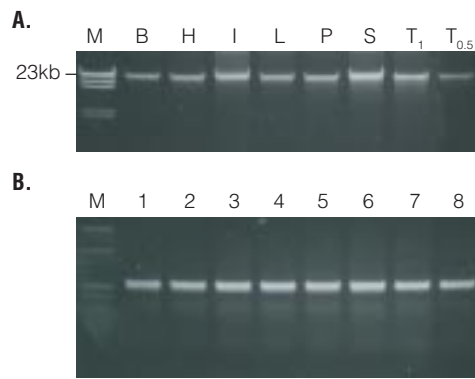


Figure 3. Isolation and analysis of DNA from whole tissues. Panel A. DNA was isolated from dissected tissue by placing the sample directly into a Reagent Cartridge from the Maxwell™ 16 Tissue DNA Purification Kit, (Cat.# AS1030). Tissues included 50mg of brain (B), heart (H), intestine (I), liver (L), pancreas (P), spleen (S) and mouse tail (T₁ is a 1cm tail snip, T_{0.5} an 0.5cm snip). Aliquots (5μl) of each sample were analyzed by agarose gel electrophoresis. Lane M, Lambda DNA/Hind III Markers (Cat.# G1711). **Panel B.** Aliquots (1μl) of each sample were amplified using a primer pair specific for a portion of the IL-1β gene and analyzed by gel electrophoresis. Lane M, 1kb DNA Ladder (Cat.# G5711).

DNA can be isolated directly from cells using the Reagent Cartridges supplied with the Maxwell™ 16 Cell DNA Purification Kit (Cat.# AS1020). The isolation format is similar to that for the other Maxwell™ 16 Reagent Cartridges, and gDNA can be isolated from cells grown either in suspension or attached to plates. Cells are collected and processed in up to 400μl volumes, or Lysis Buffer can be added directly to plates and cellular lysates collected for processing (Figure 4). A variety of prokaryotic organisms have also been tested. Many Gram negative bacteria can be processed without pretreatment, while a simple pretreatment of Gram positive bacteria may be required for optimal recovery of gDNA (Figure 4).

To demonstrate the flexibility of the Maxwell™ 16 System, its performance was tested using alternative sample types including human lung, plant leaf, and whole *Drosophila* (Figure 5, Panel A). The DNA obtained is easily visualized by agarose gel electrophoresis. Other sample types such as human hairs (root end) and buccal swabs can also be processed using the Maxwell™ 16 Instrument and Reagent Cartridges supplied with the Maxwell™ 16 Tissue DNA Purification Kit (Figure 5, Panel B). The gDNA isolated from these sample types performs well in downstream amplification-based applications. The demonstrated performance of the gDNA purification cartridges and the potential for using future Promega reagent kits for isolating RNA and gDNA from forensic samples using the Maxwell™ 16 Instrument generates maximum flexibility in a simple and compact footprint.

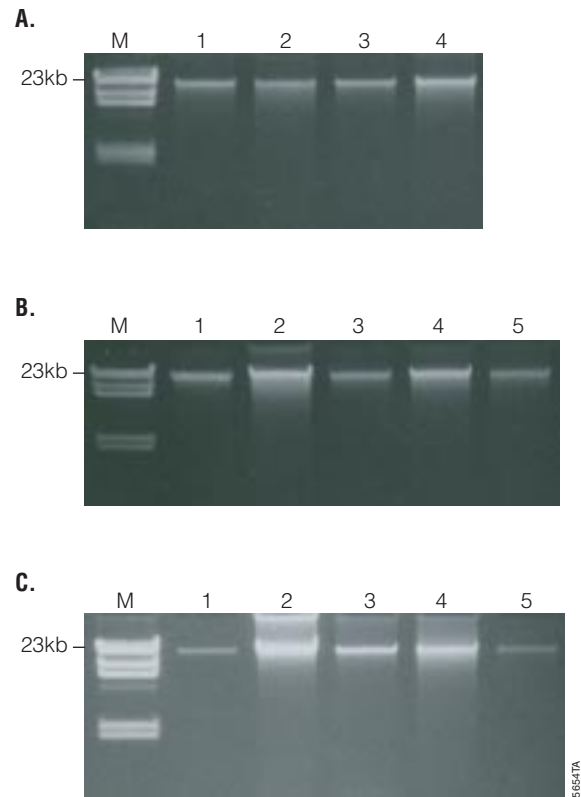


Figure 4. Isolation and analysis of gDNA from eukaryotic and prokaryotic cells. Panel A. DNA was isolated from tissue culture lines by placing samples directly into a Reagent Cartridge from the Maxwell™ 16 Cell DNA Purification Kit (Cat.# AS1020). Cell lines used (1×10^6 cells/isolation) include HeLa (lane 1), CHO (lane 2), HEK 293 (lane 3) and 3T3 (lane 4). Aliquots (5μl) of each sample were analyzed by agarose gel electrophoresis. Lane M, Lambda DNA/Hind III Markers (Cat.# G1711). **Panel B.** DNA was isolated from Gram negative bacteria by placing 400μl of overnight culture into the Reagent Cartridge supplied with the Maxwell™ 16 Cell DNA Purification Kit. Strains included *E. coli* (lane 1), *P. aeruginosa* (lane 2), *P. vulgaris* (lane 3), *S. typhimurium* (lane 4) and *Y. enterocolitica* (lane 5). Aliquots (15μl) of each sample were resolved by agarose gel electrophoresis. Lane M, Lambda DNA/Hind III Markers (Cat.# G1711). **Panel C.** DNA was isolated from Gram positive bacteria by placing 400μl of enzyme-treated culture into the Reagent Cartridge (Maxwell™ 16 Cell DNA Purification Kit). Strains included *S. aureus* (lane 1), *A. luteus* (lane 2), *B. subtilis* (lane 3), *B. cereus* (lane 4) and *E. faecalis* (lane 5). Aliquots (10μl) of each sample were analyzed by agarose gel electrophoresis. Lane M, Lambda DNA/Hind III Markers (Cat.# G1711).

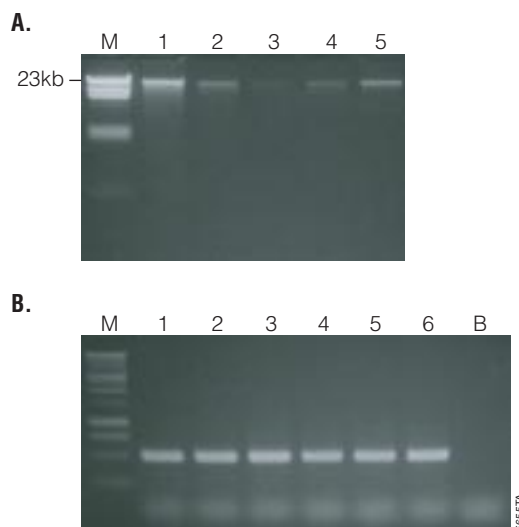


Figure 5. The Maxwell™ 16 Instrument allows a variety of samples to be processed. **Panel A.** DNA was isolated from human lung (lane 1), tomato leaf (lane 2) or 1, 3 or 5 whole *Drosophila* (lane 3–5). Lung and *Drosophila* samples were placed directly into the Maxwell™ 16 Reagent Cartridges supplied with the Maxwell™ 16 Tissue DNA Purification Kit (Cat.# AS1030); plant tissue was briefly minced by razor prior to addition to the Reagent Cartridge. Aliquots (15µl) of each sample were analyzed by agarose gel electrophoresis. Lane M, Lambda DNA/*Hind* III Markers (Cat.# G1711). **Panel B.** DNA was isolated from cotton buccal swabs (lanes 1–3) and the root end of 1, 3 or 5 human hairs (lanes 4–6). Aliquots (1µl) were amplified using primer targeting a portion of the Factor II gene and amplified products were analyzed by agarose gel. Lane M, 1kb DNA Ladder (Cat.# G5711).

Conclusion

The Maxwell™ 16 System is designed to provide labor savings and ease-of-use for low- to moderate-throughput purification of biomolecules. The system uses a compact and simple instrument that requires minimal training and maintenance, combined with prefilled reagent cartridges to improve productivity by freeing up time. The Maxwell™ 16 System can purify gDNA from whole blood (10–400µl), animal and plant tissue and eukaryotic and prokaryotic cells in culture. We have demonstrated the consistently high performance of the Maxwell™ 16 System with these sample types, while also highlighting the flexibility of the system in processing a wide variety of sample types, including whole tissues, whole organisms and buccal swabs.

Protocols

- ◆ Maxwell™ 16 Instrument Operating Manual #TM274, Promega Corporation. (www.promega.com/tbs/tm274/tm274.html)
- ◆ Maxwell™ 16 Blood DNA Purification Kit Protocol #9PIAS101, Promega Corporation. (www.promega.com/tbs/9pias101/9pias101.html)
- ◆ Maxwell™ 16 Cell DNA Purification Kit Protocol #9PIAS102, Promega Corporation. (www.promega.com/tbs/9pias102/9pias102.html)
- ◆ Maxwell™ 16 Tissue DNA Purification Kit Protocol #9PIAS103, Promega Corporation. (www.promega.com/tbs/9pias103/9pias103.html)

Ordering Information

| Product | Size | Cat.# |
|---|----------|--------|
| Maxwell™ 16 Instrument* | 1 each | AS1000 |
| Maxwell™ 16 Blood DNA Purification Kit | 48 preps | AS1010 |
| Maxwell™ 16 Cell DNA Purification Kit | 48 preps | AS1020 |
| Maxwell™ 16 Tissue DNA Purification Kit | 48 preps | AS1030 |

*For Research Use Only. Not for use in diagnostic procedures.

Related Products

| Product | Size | Cat.# |
|-------------------------------------|-------|-------|
| 1kb DNA Ladder | 500µl | G5711 |
| Lambda DNA/ <i>Hind</i> III Markers | 100µg | G1711 |

For Laboratory Use.

(a) The PCR process is covered by patents issued and applicable in certain countries*. Promega does not encourage or support the unauthorized or unlicensed use of the PCR process.

* In Europe, effective March 28, 2006, European Pat. Nos. 201,184 and 200,362 will expire. In the U.S., the patents covering the foundational PCR process expired on March 29, 2005.

Products may be covered by pending or issued patents or may have certain limitations. Please visit our Web site for more information.

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