TECHNICAL MANUAL

Maxwell® CSC Whole Blood DNA Kit

Instructions for Use of Product
AS1820

Caution: Handle cartridges with care; seal edges may be sharp.

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CE

INSTRUCTIONS FOR USE OF PRODUCT
AS1820

IVD

Revised 10/22
TM660

Maxwell® CSC Whole Blood DNA Kit

All technical literature is available at: www.promega.com/protocols/
Visit the web site to verify that you are using the most current version of this Technical Manual.
E-mail Promega Technical Services if you have questions on use of this system: techserv@promega.com

The Maxwell® CSC Whole Blood DNA Kit is only available in certain countries.

1. Description

The Maxwell® CSC Whole Blood DNA Kit[a] is used with the Maxwell® Instruments specified in Table 1 to provide an easy method for efficient, automated sample preparation and purification of genomic DNA (gDNA) from human whole blood samples. The Maxwell® CSC Instruments are designed for use with predispensed reagent cartridges and preprogrammed purification procedures, maximizing simplicity and convenience. The Maxwell® method for the CSC Whole Blood DNA Kit can process from one to the maximum number of Maxwell® CSC Instrument samples in less than 1 hour. The purified DNA can be used directly in a variety of downstream applications such as PCR.

Table 1. Supported Instruments.

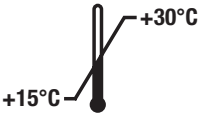
Instrument	Cat.#	Technical Manual	Maximum Sample Number
Maxwell® CSC	AS6000	TM457	16
Maxwell® CSC 48	AS8000	TM623	48

Principle of the Method: The Maxwell® CSC Whole Blood DNA Kit purifies gDNA from samples using paramagnetic particles, which provide a mobile solid phase to optimize sample capture, washing and purification of gDNA. Maxwell® Instruments are magnetic particle-handling instruments that efficiently bind nucleic acids to the paramagnetic particle in the first well of a prefilled cartridge. The samples are processed through a series of washes before the gDNA is eluted.

2. Product Components, Storage Conditions and Symbols Key

PRODUCT	SIZE	CAT.#
Maxwell® CSC Whole Blood DNA Kit	**48 preps**	**AS1820**

For In Vitro Diagnostic Use. Professional use only. Contains sufficient reagents for 48 automated isolations from 500µl of whole blood samples. Cartridges are for single use only.



Includes:

- 48 Maxwell® CSC Cartridges (CSCH)
- 50 CSC/RSC Plungers
- 50 Elution Tubes (0.5ml)
- 20ml Elution Buffer

Storage Conditions: Store the Maxwell® CSC Whole Blood DNA Kit at +15°C to +30°C.

Safety Information: The Maxwell® CSC Cartridges (CSCH) contain ethanol, isopropanol, guanidine hydrochloride and guanidine thiocyanate. Ethanol and isopropanol should be considered flammable, harmful and irritants. Guanidine thiocyanate and guanidine hydrochloride should be considered toxic, harmful and irritants. Refer to the Safety Data Sheet (SDS) for detailed safety information.

Maxwell® CSC Cartridges (CSCH) are designed to be used with potentially infectious substances. Wear appropriate protection (e.g., gloves and safety glasses) when handling infectious substances. Bleach reacts with guanidine thiocyanate, which is used in the Maxwell® CSC Cartridges (CSCH), and should not be added to any sample waste from these cartridges. Adhere to your institutional guidelines for the handling and disposal of all infectious substances when used with this system.

Caution: Handle cartridges with care; seal edges may be sharp.

Additional Information: The Maxwell® CSC Whole Blood DNA Kit components are qualified and quality-control tested to work together. It is not recommended to mix kit components between different kit lots. Use only the components provided in the kit. Do not use cartridges if the seal on the cartridge is not intact on receipt. For additional safety information, see the Safety Data Sheet, available at: **www.promega.com**.

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Symbols Key

Symbol	Explanation	Symbol	Explanation
IVD	In Vitro Diagnostic Medical Device	EC REP	Authorized Representative
+30°C +15°C	Store at +15° to +30°C.		Manufacturer
	Corrosive to skin.		Flammable
	Irritant		Caution
CE	Conformité Européenne	Σ n	Contains sufficient for "n" tests
	Warning. Pinch point hazard.		Warning. Biohazard.
LOT	Lot number	REF	Catalog number
2	Do not reuse		

3. Product Intended Purpose/Intended Use

The Maxwell® CSC Whole Blood DNA Kit is intended for use, in combination with the Maxwell® CSC Instruments and the Maxwell® CSC Whole Blood DNA purification method, as an in vitro diagnostic (IVD) medical device to perform automated isolation of genomic DNA from human whole blood samples. The purified DNA is suitable for use in amplification-based in vitro diagnostic assays.

The Maxwell® CSC Whole Blood DNA Kit is intended to be used at temperatures between 15°C and 30°C. Use outside of this temperature range can cause suboptimal results.

Whole blood samples collected in blood collection tubes containing EDTA, sodium citrate or heparin anticoagulants can be used with the Maxwell® CSC Whole Blood DNA Kit. The table below shows the acceptable time that samples can be stored under different conditions prior to use in the Maxwell® CSC Whole Blood DNA Kit.

Sample Storage Temperature	Storage Time Before Purification
15°C to 30°C	Up to 72 hours
2°C to 10°C	Up to 7 days
−65°C or lower	Indefinitely

The Maxwell® CSC Whole Blood DNA Kit is not intended for use with samples that have been collected in other types of blood collection tubes or stored outside of the conditions listed.

The Maxwell® CSC Whole Blood DNA Kit is intended for professional use only. Diagnostic results obtained using the genomic DNA purified with this system must be interpreted in conjunction with other clinical or laboratory data.

4. Product Use Limitations

The Maxwell® CSC Whole Blood DNA Kit is not intended for use with tissue samples or samples from body fluids other than human whole blood.

The Maxwell® CSC Whole Blood DNA Kit is not intended for use with nonhuman samples, including bacterial and viral samples, or for the purification of RNA.

The Maxwell® CSC Whole Blood DNA Kit performance has been evaluated by isolating DNA from 500µl whole blood samples with a white blood cell (WBC) count ranging from 4×10^6 to 1.1×10^7 cells/ml whole blood and eluting the DNA in 60µl.

The user is responsible for establishing performance characteristics necessary for downstream diagnostic applications. Appropriate controls must be included in any downstream diagnostic applications using genomic DNA purified using the Maxwell® CSC Whole Blood DNA Kit.

5. Before You Begin

Materials to Be Supplied by the User

- rotating tube mixer for liquid blood samples
- pipettors and pipette tips for sample transfer into prefilled reagent cartridges

5.A. Preparing Whole Blood Samples

The total yield of genomic DNA from whole blood samples depends on the sample white blood cell count. Each cartridge supplied in the Maxwell® CSC Whole Blood DNA Kit is designed to purify genomic DNA from 500µl of whole blood with white blood cells in the range of 4×10^6 to 1.1×10^7 cells/ml whole blood (values for a normal healthy adult; 1). We recommend performing a white blood cell count on each sample prior to DNA purification to ensure the sample falls within this range. Samples outside of this range may not provide optimal results.

Note: This kit has been tested with human whole blood samples collected in EDTA, sodium citrate and heparin tubes. Kit performance cannot be guaranteed with other types of blood collection tubes. Blood samples may be fresh (stored at 15° to 30°C for up to 72 hours), refrigerated (stored at 2°C to 10°C for up to 7 days) or frozen (stored at −65°C or lower) prior to DNA purification. Frozen samples must be thawed before processing. All blood samples must be thoroughly mixed before use.

5.B. Preparing the Maxwell® CSC Whole Blood DNA Cartridge

1. Change gloves before handling cartridges, CSC/RSC Plungers and Elution Tubes (0.5ml). Cartridges are set up in the deck tray(s) outside of the instrument before transferring the deck tray(s) containing the cartridges and samples to the instrument for purification. Place each cartridge in the deck tray(s) with well #1 (the largest well in the cartridge) farthest away from the elution tubes (Figure 2). Press down on the cartridge to snap it into position. Ensure both cartridge ends are fully seated in the deck tray. Carefully peel back the seal so that the entire seal is removed from the top of the cartridge. Ensure that all sealing tape and any residual adhesive are removed from the cartridge.
2. Transfer 500µl of blood sample to well #1 of each cartridge (well #1 is the largest well).
3. Mix the blood sample in well #1 by pipetting 5–10 times to ensure all blood has been transferred. Mixing the sample by pipetting may improve chemistry performance. Change pipette tips between samples.
4. Place one plunger into well #8 of each cartridge.
5. Place an empty elution tube into the elution tube position for each cartridge in the deck tray(s). Add 60µl of Elution Buffer to the bottom of each elution tube.

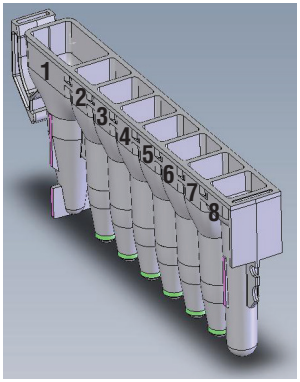
   Notes:

   a. Use only the elution tubes provided in the Maxwell® CSC Whole Blood DNA Kit. Other elution tubes may be incompatible with the Maxwell® CSC Instruments.

   b. Use only the Elution Buffer provided in the Maxwell® CSC Whole Blood DNA Kit. Other elution buffers may affect DNA purification performance.
6. Proceed to Section 6, Maxwell® Instrument Run.

Maxwell® CSC Whole Blood DNA Cartridge Preparation Notes:

Specimen or reagent spills on any part of the deck tray should be cleaned with a detergent-water solution, followed by a bactericidal spray or wipe and then water. Do not use bleach on any instrument parts.



User Adds to Wells
1. Whole blood sample (500µl)
8. CSC/RSC Plunger

Figure 1. Maxwell® CSC Cartridge. Whole blood sample is added to well #1, and a plunger is added to well #8.

Figure 2. Setup and configuration of the deck tray(s). Elution Buffer is added to the elution tubes as indicated. Deck tray shown is from the Maxwell® CSC Instrument (Cat.# AS6000).

6. Maxwell® Instrument Run

For detailed information, refer to the Technical Manual specific to your Maxwell® CSC Instrument. See Table 1.

1. Turn on the Maxwell® Instrument and Tablet PC. Log in to the Tablet PC, and start the Maxwell® IVD-mode software by double-touching the icon on the desktop. The instrument will proceed through a self-check and home all moving parts.
2. Touch **Start** on the 'Home' screen.
3. Scan or enter the bar code in the upper right corner of the Maxwell® CSC Whole Blood DNA Kit label and touch **OK** to automatically select the method to be run (Figure 3).

   Note: The Maxwell® CSC Whole Blood DNA Kit method bar code is required for DNA purification on the Maxwell® CSC Instruments. The kit label contains two bar codes. The method bar code is indicated in Figure 3. If the bar code cannot be scanned, contact Promega Technical Services.

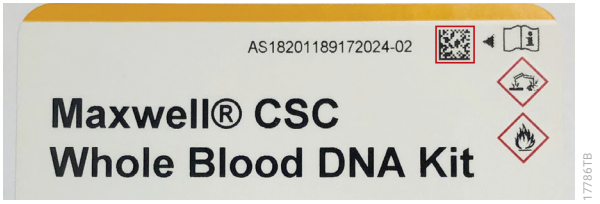


Figure 3. Kit label indicating the bar code to scan. Scan the bar code shown in the red box, upper right of the kit label, to start a purification run.

4. On the 'Cartridge Setup' screen, touch the cartridge positions to select or deselect any positions to be used for this extraction run. Enter any required sample tracking information, and touch the **Proceed** button to continue.

   Note: When using the Maxwell® CSC 48 Instrument, touch the **Front** or **Back** button to select or deselect cartridge positions for the appropriate deck tray.
5. After the door has been opened, confirm that all Extraction Checklist items have been performed. Verify that samples were added to well #1 of the cartridges, cartridges are loaded on the instrument, uncapped elution tubes are present with Elution Buffer and plungers are in well #8. Transfer the deck tray(s) containing the prepared cartridges onto the Maxwell® Instrument platform.

   Inserting the Maxwell® deck tray(s): Hold the deck tray by the sides to avoid dislodging cartridges from the deck tray. Ensure that the deck tray is placed in the Maxwell® Instrument with the elution tubes closest to the door. Angle the back of the deck tray downward and place into the instrument so that the back of the deck tray is against the back of the instrument platform. Press down on the front of the deck tray to firmly seat the deck tray on the instrument platform. If you have difficulty fitting the deck tray on the platform, check that the deck tray is in the correct orientation. Ensure the deck tray is level on the instrument platform and fully seated.

   Note: Check the identifier on 24-position Maxwell® deck trays to determine whether they should be placed in the front or back of the instrument.

6. Touch the **Start** button to begin the extraction run. The platform will retract, and the door will close.

Warning: Pinch point hazard.

Note: If using a 48-position Maxwell® Instrument and the Vision System has been enabled, the deck trays will be scanned as the platform retracts. Any errors in deck tray setup (e.g., plungers not in well #8, elution tubes not present and open) will cause the software to return to the 'Cartridge Setup' screen and problem positions will be marked with an exclamation point in a red circle. Touch the exclamation point for a description of the error and resolve all error states. Touch the **Start** button again to repeat deck tray scanning and begin the extraction run.

7. The Maxwell® Instrument will immediately begin the purification run. The screen will display the steps being performed and the approximate time remaining in the run.

   Notes:

   a. Touching the **Abort** button will abandon the run. All samples from an aborted run will be lost.

   b. If the run is abandoned before completion, you may be prompted to check whether plungers are still loaded on the plunger bar. If plungers are present on the plunger bar, you should perform **Clean Up** when requested. If plungers are not present on the plunger bar, you can choose to skip **Clean Up** when requested. The samples will be lost.

8. When the run is complete, the user interface will display a message that the method has ended.

End of Run

9. Follow on-screen instructions at the end of the method to open the door. Verify that plungers are located in well #8 of the cartridge at the end of the run. If plungers have not been removed from the plunger bar, follow the instructions in the Technical Manual appropriate to your Maxwell® Instrument (see Table 1) to perform a **Clean Up** process to attempt to unload the plungers.

10. Remove the deck tray(s) from the instrument immediately following the run to prevent evaporation of the eluates. Remove elution tubes containing DNA, and cap the tubes.

    Note: Following the automated purification procedure, the deck tray(s) will be warm. To remove a deck tray from the instrument platform, hold onto the deck tray by its sides.

    Ensure samples are removed from the instrument before running a UV sanitation protocol to avoid damage to the nucleic acid.

11. Remove the cartridges and plungers from the Maxwell® deck tray(s). Discard as hazardous waste according to your institution's procedures. Do not reuse Maxwell® CSC Cartridges, CSC/RSC Plungers or Elution Tubes.

7. Analytical Performance Evaluation

Analytical performance was evaluated using human whole blood samples with the Maxwell® CSC Whole Blood DNA Kit and Maxwell® CSC and Maxwell® CSC 48 Instruments.

7.A. DNA Yield

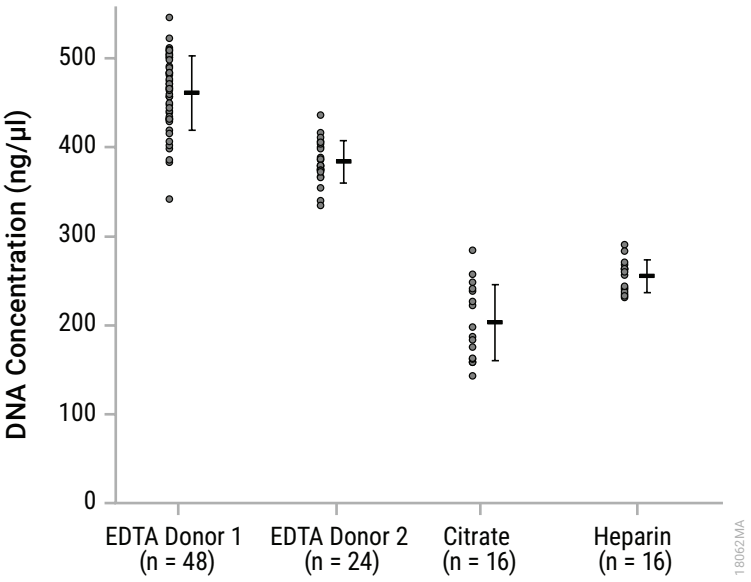


Figure 4. DNA concentration measured by absorbance (A_{260}). DNA replicates were extracted from 500µl of whole blood collected in anticoagulant tubes listed. For each data set, dots on the left represent individual measurements, while the mean DNA concentration across all replicates with standard deviation is shown on the right. Average DNA concentrations were in the range of 203.0–461.0ng/µl.

7.B. DNA Quality

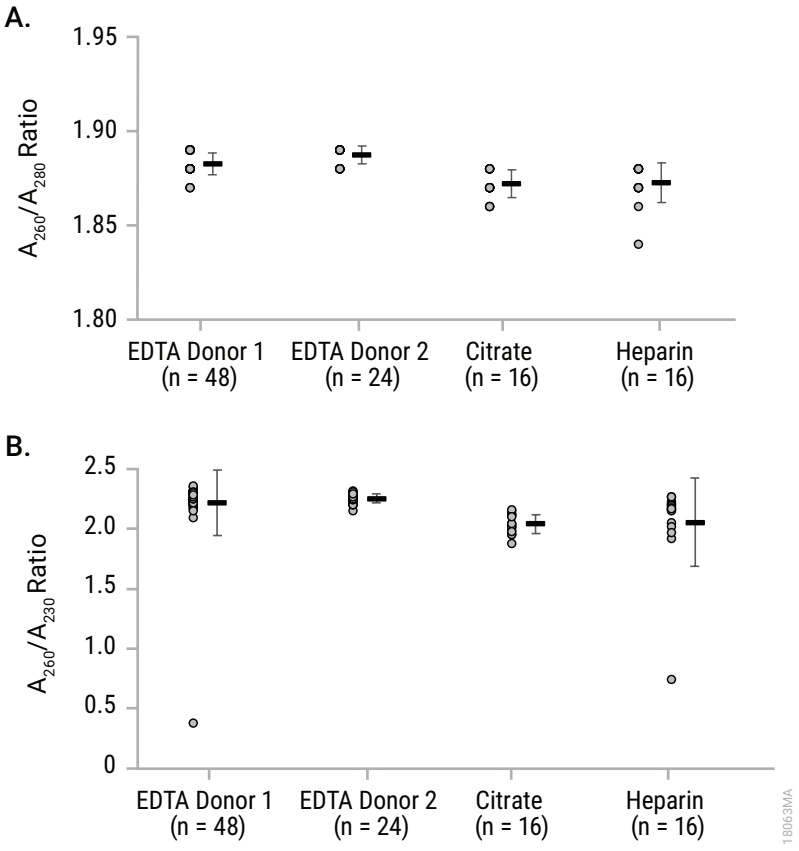


Figure 5. A_{260}/A_{280} and A_{260}/A_{230} purity ratios. DNA quality was determined by calculating absorbance ratios for eluates obtained from multiple 500µl replicates of whole blood samples collected in collection tubes containing EDTA, citrate or heparin as the anticoagulant. The figure shows A_{260}/A_{280} (**Panel A**) and A_{260}/A_{230} (**Panel B**) ratios. For each data set the dots on the left represent individual measurements, while the mean ratio with standard deviation is shown on the right. The average A_{260}/A_{280} ratios were in the range of 1.87–1.89. Average A_{260}/A_{230} ratios were in the range of 2.04–2.25.

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7.C. Reproducibility

Table 2. Reproducibility of DNA Yield. The inter-run percent coefficient of variation (% CV) for the yield of DNA purified from 3 runs of 8 replicates of whole blood (500µl each) using the Maxwell® CSC Whole Blood DNA Kit performed on Maxwell® instruments #1 and #2. The average DNA yield was evaluated by absorbance spectroscopy.

Instrument Number (n = 24)	Average Yield (µg)	Standard Deviation (µg)	% CV
1	17.8	1.9	10.7
2	16.7	1.4	8.5

Table 3. The Intra-Run Percent Coefficient of Variation. Intra-run variability was determined for multiple instrument runs covering each type of blood collection tube. Each run included 8 replicate 500µl samples of whole blood. The average yield of purified DNA from 8 replicates in each run, as evaluated by absorbance spectroscopy, standard deviation and percent coefficient of variation (% CV) are shown in the table below. Percent CV values for the runs ranged from 2.7–11.6.

Run Number (n = 8)	Blood Tube Type	Average Yield (µg)	Standard Deviation (µg)	% CV
1	EDTA[1]	18.8	1.4	7.3
2	EDTA[1]	18.0	1.9	10.3
3	EDTA[1]	16.5	1.9	11.6
4	Heparin	10.1	0.3	2.7
5	EDTA[2]	14.8	0.8	5.5
6	Citrate	9.3	0.9	9.4
7	EDTA[1]	17.0	1.5	9.0
8	EDTA[1]	17.3	1.2	7.2
9	EDTA[1]	15.9	1.2	7.7
10	EDTA[2]	16.1	0.7	4.1
11	EDTA[2]	14.9	0.6	4.3
12	Citrate	6.6	0.6	9.0
13	Heparin	9.2	0.5	5.4

[1]Donor 1
[2]Donor 2

7.D. Amplifiability

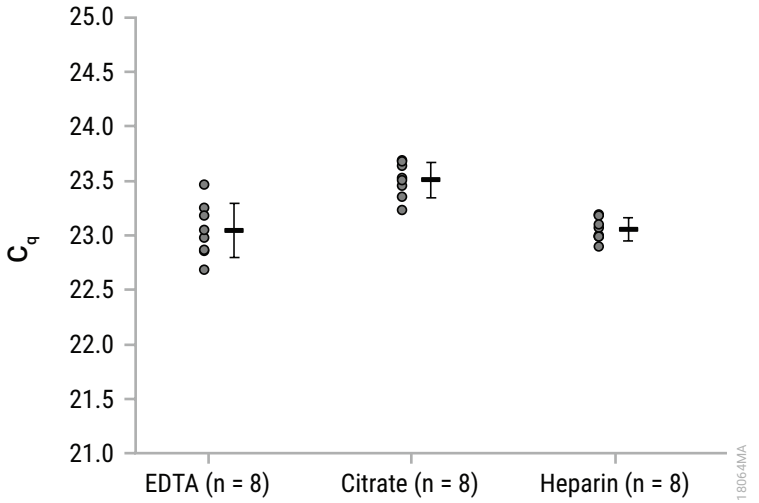

Figure 6. qPCR amplification. Eluates were analyzed by qPCR after diluting to a concentration within the qPCR standard curve. EDTA eluates were diluted 50-fold, while citrate and heparin eluates were diluted 20-fold. These samples yielded autosomal C_q values in the range of 22.69–23.69 cycles, which were below the average C_q value for the 0.0032ng/µl DNA standard (35.12 cycles).

7.E. Inhibition (Interfering Substances)

Table 4. DNA eluates were analyzed by qPCR to identify potential inhibition from interfering substances. DNA eluates were diluted to two concentrations within the qPCR standard curve that represent an eightfold difference in DNA concentration and analyzed by qPCR to determine the difference in C_q values (ΔC_q) obtained between the two dilutions. The resulting ΔC_q value is expected to be 3 ± 1 cycles based on this dilution factor. The sample eluates resulted in a ΔC_q of 2.5–3.3 cycles, indicating no detectable inhibition of DNA amplification.

Sample Number	Initial C_q	C_q of Eightfold Dilution	ΔC_q
1	23.3	26.1	2.8
2	23.1	26.0	2.9
3	22.9	25.6	2.7
4	23.5	26.1	2.5
5	23.6	26.5	3.0
6	23.2	26.4	3.2
7	23.1	26.1	3.0
8	23.1	26.2	3.1
9	23.3	26.0	2.8
10	22.9	25.8	2.9
11	22.7	25.5	2.8
12	23.0	25.9	2.9
13	23.5	26.3	2.8
14	23.1	26.0	3.0
15	22.9	26.0	3.1
16	23.2	26.5	3.3

7.F. Cross Contamination

Male and female whole blood samples, 500µl each, were processed in alternating deck positions on the Maxwell® CSC Instrument in IVD mode. The resulting eluates were analyzed using qPCR of a Y chromosome DNA target to detect any cross contamination of male DNA in the female samples. No cross contamination was detected. The female DNA samples had no detectable C_q values for the Y chromosome DNA target.

8. Clinical Performance Evaluation

Clinical performance was evaluated using human whole blood samples with the Maxwell® CSC Whole Blood DNA Kit and Maxwell® CSC 48 Instrument.

Table 5. HLA-B27 Assay with Human Whole Blood. An HLA-B27 assay was performed by an external laboratory using DNA extracted from human whole blood from 12 presumed positive and 12 presumed negative samples. DNA extraction was performed by two testers using the Maxwell® CSC Whole Blood DNA Kit and by the laboratory's standard DNA extraction method (Laboratory Reference Method). DNA eluates obtained using the Maxwell® CSC Whole Blood DNA Kit were diluted fivefold and run in a qPCR assay with the DNA eluates from the laboratory reference extraction method. The 24 samples demonstrated concordance between the two Maxwell® CSC system testers and the laboratory reference method.

Unique Patient Samples and Presumed HLA-B27 Status	Maxwell® CSC Purification System		Laboratory Reference Method
	Tester A	Tester B	
12 Positive	12 Positive	12 Positive	12 Positive
12 Negative	12 Negative	12 Negative	12 Negative

9. Troubleshooting

For questions not addressed here, please contact your local Promega Branch Office or Distributor. Contact information available at: **www.promega.com**. E-mail: **techserv@promega.com**

Symptoms	Causes and Comments
Lower than expected concentration	Blood that has undergone multiple freeze-thaw cycles may have degraded DNA. Use samples that have been collected and stored under the conditions listed in Section 3.
	Whole blood sample contained low white blood cell count. The genomic DNA concentration from blood samples depends on the number of white blood cells present in the sample.
	Whole blood sample was not mixed before processing. Be sure to mix whole blood samples before processing to ensure that the white blood cells are in suspension.
Lower than expected purity	Blood has been stored unfrozen for an extended period of time or has undergone multiple freeze-thaw cycles. Use samples that have been collected and stored under the conditions listed in Section 3.
	Blood was not mixed into the contents of well #1 when added to the cartridge. Mix the blood sample in well #1 by pipetting 5–10 times.
Resin carryover	Resin carryover is normal and does not affect downstream performance. If necessary, use an Elution Magnet ([Cat.# AS4017, Cat.# AS4018 or both]) to transfer the eluate into a new tube. See Section 11, Related Products.

Any serious incident that occurred in relation to the device that led to, or might lead to, death or serious injury of a user or patient should be immediately reported to the manufacturer. Users based in the European Union should also report any serious incidents to the Competent Authority of the Member State in which the user and/or the patient is established.

10. Reference

1. Henry, J.B. (2001) *Clinical Diagnosis and Management by Laboratory Methods*, 20th ed., W.B. Saunders Company, 509.

11. Related Products

Instrument and Accessories

Product	Size	Cat.#
Maxwell® CSC 48 Instrument*	1 each	AS8000
Maxwell® CSC Instrument*	1 each	AS6000
Maxwell® RSC/CSC Deck Tray	1 each	SP6019
Maxwell® RSC/CSC 48 Front Deck Tray	1 each	AS8401
Maxwell® RSC/CSC 48 Back Deck Tray	1 each	AS8402
Elution Tubes (0.5ml)	50/pack	AS6201
Elution Magnet, 16 Position	1 each	AS4017
Elution Magnet, 24 Position	1 each	AS4018

*For In Vitro Diagnostic Use. This product is only available in certain countries.

Maxwell® CSC Reagent Kits

Visit **www.promega.com** for a list of available Maxwell® CSC purification kits.

12. Summary of Changes

The following changes were made to the 10/22 revision of this document:

1. Section 3 was renamed to Product Intended Purpose/Intended Use.
2. Sections 7 and 8 were added and subsequent sections renumbered.
3. Document updated for compliance with Regulation (EU) 2017/746 on in vitro diagnostic medical devices.
4. Disclaimer was added.

[a]U.S. Pat. No. 7,329,488 and S. Korean Pat. No. 100483684.