TECHNICAL MANUAL

# DNA IQ<sup>™</sup> Casework Pro Kit for Maxwell<sup>®</sup> 16

Instructions for Use of Products AS1240 and DC6745

Promega



Revised 12/16 TM332

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# **DNA IQ<sup>TM</sup> Casework Pro Kit** for Maxwell<sup>®</sup> 16

	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	
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## 1. Description

The DNA IQ<sup>™</sup> Casework Pro Kit for Maxwell<sup>®</sup> 16<sup>(a)</sup> is used with the Maxwell<sup>®</sup> 16 Instrument (Cat.# AS2000, Cat.# AS3060) configured for low elution volume (LEV) and is specifically designed for optimal DNA extraction from forensic casework samples. These samples include blood stains, semen stains, hair, cigarette butts, tissue samples and "touch" DNA samples regularly encountered in forensic DNA analysis.

The Casework Extraction Kit was developed to improve DNA extraction efficiency from a broad panel of sample types (Figure 1). This protocol describes the use of the Casework Extraction Kit (Cat.# DC6745) for preprocessing samples before DNA extraction with the DNA IQ<sup>™</sup> Casework Pro Kit for Maxwell<sup>®</sup> 16.



# Figure 1. DNA was extracted from mock casework sample types using the Incubation Buffer (from the Tissue and Hair Extraction Kit, Cat.# DC6740) or Casework Extraction Buffer and isolated using the DNA IQ<sup>™</sup> Casework Pro Kit for Maxwell<sup>®</sup> 16.

Extraction from certain sample types (e.g., sperm cells) traditionally requires the use of dithiothreitol (DTT) as part of the extraction method. The Casework Extraction Kit includes 1-Thioglycerol, which, unlike DTT, can be stored at  $2-10^{\circ}$ C, and provides extraction performance comparable to that with DTT. This Technical Manual provides instructions to use 1-Thioglycerol for sample preprocessing.

The Maxwell<sup>®</sup> 16 LEV Instrument is supplied with preprogrammed DNA purification procedures, uses prefilled reagent cartridges, and elutes DNA samples in small volumes. The instrument can process up to 16 samples in approximately 30 minutes.

The DNA IQ<sup>™</sup> Casework Pro Kit for Maxwell<sup>®</sup> 16 uses the DNA IQ<sup>™</sup> Resin to purify DNA, maximizing DNA yield and purity for use in STR analysis. The Maxwell<sup>®</sup> 16 Instrument is a magnetic-particle-handling instrument that efficiently transports the DNA IQ<sup>™</sup> Resin through purification reagents in prefilled cartridges (Figure 2), mixing the resin with the reagents during processing. The paramagnetic particle-based methodology avoids common problems experienced with other automated systems, such as clogged tips or partial reagent transfers, which can lead to suboptimal DNA purification.

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#### 2. Product Components and Storage Conditions

PRODUCT	SIZE	CAT.#	
DNA IQ™ Casework Pro Kit for Maxwell® 16	48 preps	AS1240	
Not For Medical Diagnostic Use. Sufficient for 48 automated iso	lations from forensic casework s	amples.	
Includes:			

• 48 Maxwell<sup>®</sup> 16 LEV Cartridges (MCB)

- 50 LEV Plungers
- 50 Elution Tubes, 0.5ml
- 20ml Elution Buffer
- 32ml Lysis Buffer

#### **Purchase Separately**

PRODUCT	SIZE	CAT.#	
Casework Extraction Kit	100 reactions	DC6745	
Not For Medical Diagnostic Use. Sufficient for 100 reactions	s of 400µl each. Includes:		

• 50ml Casework Extraction Buffer

- 2 × 10mg Proteinase K
- 900µl 1-Thioglycerol
- 1.25ml Nuclease-Free Water

**Storage Conditions:** Store the DNA IQ<sup>™</sup> Casework Pro Kit for Maxwell<sup>®</sup> 16 and Casework Extraction Buffer at 15–30°C. Store 1-Thioglycerol at 2–10°C. Store Proteinase K at –30 to –10°C. Store Nuclease-Free Water below 30°C.

**Safety Information:** The reagent cartridges contain flammable substances ethanol and isopropanol, and the harmful substance guanidine thiocyanate. 1-Thioglycerol is a toxic substance. We highly recommend the use of gloves and aerosol-resistant pipette tips.







#### 3. Maxwell® 16 Instrument Hardware and Firmware Setup

The Maxwell<sup>®</sup> 16 Instrument (Cat.# AS2000) or Maxwell<sup>®</sup> 16 Forensic Instrument (Cat.# AS3060) configured for low volume elution (LEV) is required for use with the DNA IQ<sup>™</sup> Casework Pro Kit for Maxwell<sup>®</sup> 16. Users with a Maxwell<sup>®</sup> 16 Instrument configured for standard elution volume (SEV) need to reconfigure the instrument using the Maxwell<sup>®</sup> 16 LEV Hardware Kit (Cat.# AS1250). Reconfiguring the instrument is easy. For instructions to set up the instrument, please refer to the *Maxwell<sup>®</sup> 16 Instrument Technical Manual* #TM295 for Cat.# AS2000 or the *Maxwell<sup>®</sup> 16 Forensic Instrument Technical Manual* #TM321 for Cat.# AS3060.

The first time that the Maxwell<sup>®</sup> 16 Instrument is powered up, a series of user prompts will appear on the Navigation LCD. The DNA IQ<sup>™</sup> Casework Pro Kit for Maxwell<sup>®</sup> 16 is intended for use with the LEV (low elution volume) settings and the Forensic method on the instrument. Once the Forensic method is set up on the instrument, all subsequent power-ups of the instrument will automatically default to these settings.

# 4. Sample Preprocessing

Depending on the sample type, there are different preprocessing steps that must be performed prior to running the automated method for the Maxwell<sup>®</sup> 16 Instrument.

To maximize the amount of DNA purified from forensic casework materials, use the protocol appropriate for your sample type. Protocols include:

- 1. Samples on a Solid Support (Section 4.A)
- 2. Liquid Samples (Section 4.B)
- 3. Differential Extraction Samples (Section 4.C).

All protocols involve a Proteinase K treatment, which is required to maximize recovery and yield from a variety of sample types, including small amounts of sample on a solid matrix, such as a swab or fabric. DNA samples extracted using a Proteinase K treatment generally exhibit better locus-to-locus balance in downstream STR analysis.

**Note:** These preprocessing protocols use the Casework Extraction Buffer included in the Casework Extraction Kit (Cat.# DC6745) for the Proteinase K digestion. If another proteinase K digestion buffer is used for comparison, the concentration of SDS must be below 0.5%, or a precipitate may form when the Lysis Buffer is added.

#### 4.A. Samples on a Solid Support

This preprocessing protocol allows optimal DNA extraction from samples on a solid support, such as swabs or fabric, using a Proteinase K incubation. Two protocols are provided below (Options 1 and 2), depending on which spin baskets and microtubes are selected.

#### Materials to Be Supplied by the User

- 56°C heat block or water bath
- Casework Extraction Kit (Cat.# DC6745)
- aerosol-resistant pipette tips
- Preprocessing Spin Baskets and Tubes.
  - 1. DNA IQ<sup>™</sup> Spin Baskets (Cat.# V1225) with ClickFit MicroTubes, 1.5ml (Cat.# V4745)
  - 2. CW Spin Baskets (Cat.# AS8101) with CW Microfuge Tubes, 1.5ml (Cat.# AS8201)

# Preparation of Stock Proteinase K Solution for Sample Preprocessing

Add 556µl of Nuclease-Free Water to one tube of lyophilized Proteinase K, and gently invert to dissolve. The final concentration of Proteinase K will be 18mg/ml. Store Proteinase K Solution at –20°C.

# Option 1: DNA IQ<sup>™</sup> Spin Baskets (Cat.# V1225) with ClickFit MicroTubes, 1.5ml (Cat.# V4745)

- 1. Place solid substrate (e.g., fabric or swab head) at the bottom of a labeled ClickFit Microtube.
- 2. Create the Extraction Mix by adding the final volume of each reagent listed in Table 1 to a clean tube.

# Table 1. Extraction Mix for Solid Support Samples with DNA IQ<sup>™</sup> Spin Baskets.

	<b>Volume Per</b>		Number of		
<b>Extraction Mix Component</b>	Extraction	×	Extractions	=	<b>Final Volume</b>
Casework Extraction Buffer	386µl	×		=	
Proteinase K (18mg/ml)	10µl	×		=	
1-Thioglycerol	4µl	×		=	
Total Reaction Volume	400µl	×		=	

Note: 1-Thioglycerol is viscous. Pipet slowly.

- 3. Briefly vortex the Extraction Mix, and dispense 400µl to each ClickFit Microtube containing solid substrate.
- 4. Close the tube lid, vortex sample at high speed for 5 seconds, and incubate the sample at 56°C for 30 minutes.
- 5. Place a DNA IQ<sup>™</sup> Spin Basket into a clean, labeled ClickFit Microtube. Transfer the sample to the DNA IQ<sup>™</sup> Spin Basket with forceps, being sure to orient the swab or fabric toward the bottom of the spin basket. Transfer the lysate from the incubation tube to the spin basket, and close the tube.
- 6. Centrifuge at room temperature for 2 minutes at maximum speed in a microcentrifuge. Carefully remove the DNA IQ<sup>™</sup> Spin Basket.



#### 4.A. Samples on a Solid Support (continued)

- 7. Add 200µl of Lysis Buffer to the tube containing extract.
- 8. Close the lid of the tube, and vortex the sample for 5-10 seconds.
- 9. The sample is now ready for automated DNA extraction using the Maxwell<sup>®</sup> 16 LEV Instrument. Proceed to Section 5 for cartridge preparation and instrument setup.
   Note: Store preprocessed sample at room temperature (15–30°C) overnight, if necessary.

#### Option 2: CW Spin Baskets (Cat.# AS8101) with CW Microfuge Tubes, 1.5ml (Cat.# AS8201)

- 1. Place a CW Spin Basket into a labeled CW Microfuge Tube.
- 2. Place solid substrate (e.g., fabric or swab head) at the bottom of a CW Spin Basket.
- 3. Create the Extraction Mix by adding the final volume of each reagent listed in Table 2 to a clean tube.

#### Table 2. Extraction Mix for Solid Support Samples with CW Spin Baskets.

	Volume Per		Number of		
<b>Extraction Mix Component</b>	Extraction	×	Extractions	=	<b>Final Volume</b>
Casework Extraction Buffer	286µl	×		=	
Proteinase K (18mg/ml)	10µl	×		=	
1-Thioglycerol	4µl	×		=	
Total Reaction Volume	300µl	×		=	

Note: 1-Thioglycerol is viscous. Pipet slowly.

- 4. Briefly vortex the Extraction Mix. Dispense 300µl to each CW Spin Basket containing solid substrate.
- 5. Close the tube lid, vortex sample at high speed for 5 seconds, and incubate at 56°C for 30 minutes.
- 6. Centrifuge at room temperature for 2 minutes at maximum speed in a microcentrifuge. Carefully remove the CW Spin Basket.
- 7. Add 200µl of Lysis Buffer to the tube containing extract.
- 8. Close the lid of the tube, and vortex the sample for 5–10 seconds.
- 9. The sample is now ready for automated DNA extraction using the Maxwell<sup>®</sup> 16 LEV Instrument. Proceed to Section 5 for cartridge preparation and instrument setup.

Note: Store preprocessed sample at room temperature (15–30°C) overnight, if necessary.

# 4.B. Liquid Samples

This preprocessing protocol results in optimal lysis from samples in an aqueous solution using a proteinase K treatment.

## Materials to Be Supplied by the User

- 56°C heat block or water bath
- ClickFit Microtubes, 1.5ml (Cat.# V4745) or CW Microfuge Tubes, 1.5ml (Cat.# AS8201)
- Casework Extraction Kit (Cat.# DC6745)
- aerosol-resistant pipette tips

Note: Using ClickFit Microtubes or CW Microfuge Tubes prevents the lid from opening during heated incubation.

# Preparation of Stock Proteinase K Solution for Sample Preprocessing

Add 556µl of Nuclease-Free Water to one tube of lyophilized Proteinase K, and gently invert to dissolve. The final concentration of Proteinase K will be 18mg/ml. Store Proteinase K Solution at -20°C.

#### **Extraction of Liquid Samples**

1. Pipet liquid sample into the bottom of a labeled ClickFit Microtube or CW Microfuge Tube, and add 10µl of Proteinase K Solution, 4µl of 1-Thioglycerol and Casework Extraction Buffer to a total volume of 400µl.

Do not exceed a final volume of  $400\mu$ l.

Note: 1-Thioglycerol is viscous. Pipet slowly.

- 2. Close the tube lid, vortex sample at high speed for 5 seconds, and incubate the sample at 56°C for 30 minutes.
- 3. Add 200µl of Lysis Buffer to each sample.
- 4. Close the lid of the tube, and vortex the sample for 5–10 seconds.
- 5. The sample is now ready for automated DNA extraction using the Maxwell<sup>®</sup> 16 LEV Instrument. Proceed to Section 5 for cartridge preparation and instrument setup.

Note: Store preprocessed sample at room temperature (15–30°C) overnight, if necessary.



## 4.C. Differential Extraction Samples

Differential extraction is a method for separating a forensic sample into two fractions: sperm and epithelial. Epithelial cells are lysed using proteinase K in the absence of a reducing agent (e.g., DTT) to prevent the lysis of spermatozoa. The Differex<sup>™</sup> System (Cat.# DC6801, DC6800) can be used to quickly and efficiently separate male and female sample fractions. See the *Differex<sup>™</sup> System Technical Manual* #TM331 for a protocol to separate sperm and epithelial fractions, available at: www.promega.com/protocols

## Materials to Be Supplied by the User

- 56°C heat block or water bath
- ClickFit Microtubes, 1.5ml (Cat.# V4745) or CW Microfuge Tubes, 1.5ml (Cat.# AS8201)
- Casework Extraction Kit (Cat.# DC6745)
- aerosol-resistant pipette tips

Note: Using ClickFit Microtubes or CW Microfuge Tubes prevents the lid from opening during heated incubation.

# Preparation of Stock Proteinase K Solution for Sample Preprocessing

Add 556µl of Nuclease-Free Water to one tube of lyophilized Proteinase K, and gently invert to dissolve. The final concentration of Proteinase K will be 18mg/ml. Store Proteinase K Solution at -20°C.

#### **DNA Extraction from Sperm Fraction**

- To the sperm fraction, add 10µl of Proteinase K Solution, 4µl of 1-Thioglycerol and Casework Extraction Buffer to a total volume of 400µl. Do not exceed a final volume of 400µl.
   Note: 1-Thioglycerol is viscous. Pipet slowly.
- 2. Close the tube lid, vortex sample at high speed for 5 seconds, and incubate the sample at 56°C for 30 minutes.
- 3. Add 200µl of Lysis Buffer to each sample.
- 4. Close the lid of the tube, and vortex the sample for 5-10 seconds.
- The sample is now ready for automated DNA extraction using the Maxwell<sup>®</sup> 16 LEV Instrument. Proceed to Section 5 for cartridge preparation and instrument setup.
   Note: Store preprocessed sample at room temperature (15–30°C) overnight, if necessary.

#### DNA Extraction from Epithelial Fraction

- 1. To an epithelial fraction of up to 400µl, add 200µl of Lysis Buffer.
- 2. Close the lid of the tube, and vortex the sample for 5–10 seconds.
- 3. The sample is now ready for automated DNA extraction using the Maxwell® 16 LEV Instrument. Proceed to Section 5 for cartridge preparation and instrument setup.

Note: Store preprocessed sample at room temperature (15–30°C) overnight, if necessary.

#### 5. Maxwell<sup>®</sup> 16 Automated DNA Purification Protocol

#### 5.A. Preparation of Samples for Maxwell® 16 LEV Cartridges

1. Change gloves before handling cartridges, LEV Plungers and Elution Tubes. Place the cartridges to be used in the Maxwell<sup>®</sup> 16 LEV Cartridge Rack (Cat.# AS1251). Place each cartridge in the rack with the label side facing away from the Elution Tubes. Press down on the cartridge to snap it into position. Carefully peel back the seal so that all plastic comes off the top of the cartridge. Ensure that all sealing tape and any residual adhesive are removed before placing cartridges in the instrument.

#### Notes:

- 1. If you are processing fewer than 16 samples, center the cartridges on the platform.
- 2. Clean sample or reagent spills on any part of the Maxwell<sup>®</sup> 16 LEV Cartridge Rack with a detergent-water solution, followed by 70% ethanol, then water. Do not use bleach on any instrument parts.
- 2. Rotate the cartridge rack 180 degrees such that well #1 is closest to you. Add sample lysate to well #1. Repeat for all samples.

**Note:** The order of samples is now 16–1, left to right. Ensure your samples are added in the correct order.



3. Change gloves and rotate the rack 180 degrees again so the well #1 is furthest from you, and place an LEV Plunger in well #8 of each cartridge. Well #8 is the well closest to the Elution Tube.



## 5.A. Preparation of Samples for Maxwell® 16 LEV Cartridges (continued)

4. Place Elution Tubes in the front of the Maxwell<sup>®</sup> 16 LEV Cartridge Rack. Add 50µl of Elution Buffer to the bottom of each Elution Tube.



#### Notes:

- 1. Ensure that the Elution Buffer is in the bottom of the tube. If Elution Buffer is on the side of the tube, the elution may be suboptimal.
- 2. Use only the Elution Tubes provided with the kit; other tubes may not work with the Maxwell® 16 Instrument.
- 3. Use only the Elution Buffer supplied with the DNA IQ<sup>™</sup> Casework Pro Kit for Maxwell<sup>®</sup> 16; other buffers can negatively affect downstream analysis (e.g., poor-quality STR profiles).
- 5. Proceed to Section 5.B for Cat.# AS2000 instruments or Section 5.C for Cat.# AS3060 instruments.

#### 5.B. Setup for AS2000 Maxwell® 16 Instruments

Refer to the Maxwell® 16 Instrument Operating Manual #TM295 for more detailed information.

To run the "Casework" protocol, you must have Maxwell® 16 firmware version 4.0 or higher installed on your instrument.

- 1. Turn on the Maxwell<sup>®</sup> 16 Instrument. The instrument will power up, display the firmware version number, proceed through a self-check and home all moving parts.
- 2. Verify that the instrument settings indicate an "LEV" hardware configuration and "Fnsc" operational mode setting.
- 3. Select "Run" on the Menu screen, and press the Run/Stop button to start the method.
- 4. Select "DNA" on the Menu screen, then select "OK" at the Verification screen.



Warning: Pinch point hazard.

6. Transfer the Maxwell<sup>®</sup> 16 LEV Cartridge Rack containing the prepared cartridges to the Maxwell<sup>®</sup> 16 Instrument platform. Ensure that the rack is placed in the Maxwell<sup>®</sup> 16 Instrument with the Elution Tubes closest to the door. The rack will only fit in the instrument in this orientation. If you have difficulty fitting the rack on the platform, check that the rack is in the correct orientation. Ensure that the cartridge rack is level on the instrument platform.

Note: Hold the Maxwell® 16 LEV Cartridge Rack by the sides to avoid dislodging cartridges from the rack.

7. Verify that samples were added to well #1 of the cartridges, Elution Tubes are present with 50µl of Elution Buffer and LEV Plungers are in well #8.



Press the Run/Stop button. The platform will retract. Close the door. **Warning:** Pinch point hazard.

9. The Maxwell<sup>®</sup> 16 Instrument will immediately begin the purification run. The screen will display the steps performed and the approximate time remaining in the run.

#### Notes:

- 1. Pressing the Run/Stop button or opening the door will pause the run.
- 2. If the run is abandoned before completion, the instrument will wash the particles off the plungers and eject the plungers into well #8 of the cartridges. However, the sample DNA will remain on the particles and can be easily recovered either by replacing the plungers in well #8 and rerunning the purification, or by removing the well contents and manually finishing the isolation process as instructed in the DNA IQ<sup>™</sup> System—Small Sample Casework Protocol Technical Bulletin #TB296.
- 10. When the automated purification run is complete, the LCD screen will display a message that the method has ended.

# **End of Run**

- 11. Follow on-screen instructions at the end of the method to open the door. Verify that plungers are located in well #8 of the cartridges. If plungers are not removed from the magnetic plunger bar, push them down gently by hand to remove them.
- 12. Press the Run/Stop button to extend the platform out of the instrument.
- 13. Remove the Maxwell<sup>®</sup> 16 LEV Cartridge Rack from the instrument. Remove Elution Tubes containing DNA, and close the tubes.

**Note:** Some paramagnetic particles may be present in the Elution Tube and can be removed by capturing particles using the Maxwell<sup>®</sup> 16 LEV Magnet (Cat.# AS1261) or 0.5ml MagneSphere<sup>®</sup> Technology Magnetic Separation Stand (Cat.# Z5341) and transferring the DNA to a new tube.

14. Remove cartridges and plungers from the Maxwell<sup>®</sup> 16 LEV Cartridge Rack and discard as hazardous waste. Do not reuse reagent cartridges, LEV Plungers or Elution Tubes.

# 5.C. Setup for AS3060 Maxwell® 16 Forensic Instruments

Refer to the Maxwell® 16 Forensic Instrument Technical Manual #TM321 for detailed information.

- 1. Turn on the Maxwell<sup>®</sup> 16 Forensic Instrument. The instrument will power up, display the firmware version number, proceed through a self-check and home all moving parts.
- 2. Verify that the Home screen indicates "LEV" and the LEV hardware is present. Press "Run" to continue.
- 3. Enter user and PIN, if this option is enabled.
- 4. At the Protocols screen, select "Casework".
- 5. On the next screen, verify that the correct method and user were chosen. Select "Run/Stop" to continue.
- 6. Open the door when prompted on the screen, then select "Run/Stop".
- Warning: Pinch point hazard.
- 7. Follow on-screen instructions for bar code reader input if this option is enabled.
- 8. Transfer the Maxwell<sup>®</sup> 16 LEV Cartridge Rack containing the prepared cartridges to the Maxwell<sup>®</sup> 16 Instrument platform. Ensure that the rack is placed in the Maxwell<sup>®</sup> 16 Instrument with the Elution Tubes closest to the door. The rack will only fit in the instrument in this orientation. If you have difficulty fitting the rack on the platform, check that the rack is in the correct orientation. Ensure the rack is level on the instrument platform.

Note: Hold the Maxwell<sup>®</sup> 16 LEV Cartridge Rack by the sides to avoid dislodging cartridges from the rack.

- 9. Verify that samples were added to well #1 of the cartridges, cartridges in the rack are loaded on the instrument, Elution Tubes are present with 50µl of Elution Buffer and LEV Plungers are in well #8.
- 10. Press the Run/Stop button. The platform will retract. Close the door.



Warning: Pinch point hazard.

The Maxwell<sup>®</sup> 16 Forensic Instrument will immediately begin the purification run. The screen will display the steps performed and the approximate time remaining in the run.

#### Notes:

- 1. Pressing the Run/Stop button or opening the door will pause the run.
- 2. If the run is abandoned before completion, the instrument will wash the particles off the plungers and eject the plungers into well #8 of the cartridge. However, the sample DNA will remain on the particles and can be recovered easily either by replacing the plungers in well #8 and rerunning the purification, or by removing the well contents and manually finishing the isolation process as instructed in the DNA IQ<sup>™</sup> System—Small Sample Casework Protocol Technical Bulletin #TB296.
- 11. When the automated purification run is complete, follow instructions on the screen for data transfer. For detailed instructions, refer to the *Maxwell*<sup>®</sup> *16 Forensic Instrument Technical Manual* #TM321 and *Maxwell*<sup>®</sup> *Sample Track Software Technical Manual* #TM314.

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#### **End of Run**

- 12. Follow on-screen instructions at the end of the method to open the door. Verify that plungers are located in well #8 of the cartridges. If plungers are not removed from the magnetic plunger bar, push them down gently by hand to remove them.
- 13. Press the Run/Stop button to extend the platform out of the instrument.
- 14. Remove the Maxwell<sup>®</sup> 16 LEV Cartridge Rack from the instrument. Remove Elution Tubes containing DNA, and cap the tubes.

**Note:** Some paramagnetic particles may be present in the Elution Tube and can be removed by capturing particles using the Maxwell<sup>®</sup> 16 LEV Magnet (Cat.# AS1261) or 0.5ml MagneSphere<sup>®</sup> Technology Magnetic Separation Stand (Cat.# Z5341) and transferring the DNA to a new tube.

15. Remove cartridges and plungers from the cartridge rack, and discard as hazardous waste. Do not reuse reagent cartridges, LEV Plungers or Elution Tubes.

Note: Ensure samples are removed before the UV light treatment to avoid damage to the nucleic acid.

#### 5.D. Storing Eluted DNA

If DNA is not analyzed immediately, store the eluted DNA on ice or at 4°C for up to 24 hours. For longer term storage, consult laboratory guidelines. Freezing samples at -20°C or -70°C has been shown to preserve DNA for longer periods of time.

# 6. 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	Possible Causes and Comments		
Low DNA yield	Insufficient sample was processed. Add more starting material for preprocessing to increase yield.		
	Too much sample was processed. DNA isolation using the Maxwell <sup>®</sup> 16 LEV Cartridge is most efficient when there is ≤800µl in well #1. Larger volumes can be processed but may reduce isolation efficiency.		
	Insufficient Lysis Buffer. Dispense an equal volume of Lysis Buffer to sample volume during the binding step in well #1. A minimum of 200µl of Lysis Buffer should be dispensed into the Maxwell <sup>®</sup> <u>16 LEV Cartridge.</u>		
	Insufficient lysis of sperm cells after differential extraction. Sperm samples typically require extraction with proteinase K.		
Instrument calibration error	Verify nothing is physically blocking movement of the platform, plunger bar or magnetic rod assembly.		
	Turn the machine off then on to cycle the power. The instrument will rehome itself. If the calibration error occurs again after power cycling, contact Promega for service.		
	Turn the machine off then on to cycle the power. After cycling power, run a "Demo" method without any cartridges in the machine. If another calibration error occurs during the "Demo" run, contact Promega for service.		
	Ensure the Maxwell <sup>®</sup> 16 LEV Hardware Kit (Cat.# AS1250) is installed.		
	The cartridges were not completely seated on the platform. Ensure the cartridges are pressed firmly into place.		
	Incorrect elution tube was used. Use only the 0.5ml Elution Tube provided with the DNA IQ <sup>™</sup> Casework Pro Kit for Maxwell <sup>®</sup> 16. Other tubes may have different dimensions.		



Symptoms	Possible Causes and Comments		
Resin carryover during elution	A small amount of resin was visible in the Elution Tube. The presence of resin particles will not affect the final DNA concentration or downstream applications. If desired, an additional resin capture step may be performed using the Maxwell 16 <sup>®</sup>		
	ogy Magnetic Separation Stand (Cat.# Z5341).		
	Use the Maxwell <sup>®</sup> 16 High Strength LEV Magnetic Rod and Plunger Bar Adaptor (Cat.# SP1070) to significantly improve the recovery of resin particles at each step and reduce the amount of resin in the Elution Tube.		

#### 6. Troubleshooting (continued)

#### 7. Extraction Buffer Options

Additional extraction buffers are compatible with the DNA IQ<sup>™</sup> Casework Pro Kit for Maxwell<sup>®</sup> 16. The Casework Extraction Buffer provided as part of the Casework Extraction Kit (Cat.# DC6745) is recommended, but different laboratories have reported success with Bone Incubation Buffer.

#### **Bone Incubation Buffer**

10mM Tris 100mM NaCl 50mM EDTA 0.5% SDS

Note: Some users prefer to process reference samples in heated DNA IQ<sup>™</sup> Lysis Buffer. We do **not** recommend this method for casework samples.



#### 8. Related Products

Prod.uct	Size	Cat.#
Maxwell® 16 LEV Hardware Kit	1 each	AS1250
Maxwell® 16 LEV Cartridge Rack	1 each	AS1251
DNA IQ™ Reference Sample Kit for Maxwell® 16	48 preps	AS1040
Differex™ System	50 samples	DC6801
-	200 samples	DC6800
Proteinase K	100mg	V3021
Maxwell® 16 LEV Magnet	1 each	AS1261
Maxwell® 16 High Strength LEV Magnetic Rod and Plunger Bar Adaptor	1 each	SP1070
MagneSphere® Technology Magnetic Separation Stand (twelve-position)	1 each	Z5341
DNA IQ™ Spin Baskets	50/pack	V1225
ClickFit Microtube, 1.5ml	100/pack	V4745
-	1,000/pack	V4741
CW Spin Baskets	50/pack	AS8101
CW Microfuge Tube, 1.5ml	50/pack	AS8201

#### 9. Summary of Changes

The following changes were made to the 12/16 revision of this document:

Options for processing samples on a solid support using either DNA IQ<sup>™</sup> Spin Baskets (Cat.# V1225) with ClickFit MicroTubes (Cat.# V4741, V4745), or CW Spin Baskets (Cat.# AS8101) with CW Microfuge Tubes (Cat.# AS8201), were added to Section 4.A.

<sup>(a)</sup>U.S. Pat. Nos. 6,027,945, 6,368,800 and 6,673,631, European Pat. No. 1 204 741 and Japanese Pat. No. 4425513.

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