



Crime Prep Adem-Kit

AutoMag Solution

(Cat #06210)

Instruction manual for automation protocol

ADEMTECH SA
Parc scientifique Unitec 1
4, allée du Doyen G. Brus 33600 PESSAC

France

Tel: +33557020201
Fax +3355702020

Visit our Web site: www.ademtech.com

<i>Table of contents</i>	3
---------------------------------	----------

<i>Introduction</i>	4
----------------------------	----------

- | | |
|---|---|
| 1. Description | 4 |
| 2. Product component and storage conditions | 4 |
| 3. Equipment and reagents to be supplied | 6 |

<i>Crime Prep Adem-Kit Protocol</i>	7
--	----------

- | | |
|-----------------------------------|----|
| 1. Reagents preparation | 7 |
| 2. Perform Lysis | 8 |
| 3. Instruction for DNA extraction | 10 |

<i>Troubleshooting</i>	14
-------------------------------	-----------

<i>Warranty</i>	15
------------------------	-----------

<i>Ordering Information</i>	15
------------------------------------	-----------

Introduction

1. Description

The Crime Prep Adem-Kit is specially designed for optimal DNA extraction from forensic casework samples. The Crime Prep Adem-Kit was developed to improve DNA extraction efficiency from a broad panel of sample types. These samples include blood stains, hair, cigarette butts, tissues samples and “touch” DNA samples regularly encountered in forensic DNA analysis.

2. Product Component and storage conditions

Kit Content: Each Crime Prep Adem-Kit contains sufficient reagents to perform 96 samples using the following standard protocol.

Item	Crime Prep Adem-Kit
Cat No.	06210
Package size	1 x 96 samples
Crime Lysis Buffer	40mL
Proteinase K (10mg/ml)	5mL
LB Buffer	25 mL
Silica-MasterBeads	1.5mL
Washing Buffer I	24mL
Washing Buffer II	17mL
Elution Buffer	6mL

Crime Prep Adem-Kit (#06210)	
Reagents	Storage condition
Crime Lysis Buffer	+2-8°C
Proteinase K (10mg/ml)	+2-8°C
LB Buffer	+2-8°C
Silica-MasterBeads	+2-8°C
Washing Buffer I	+2-8°C
Washing Buffer II	+2-8°C
Elution Buffer	+2-8°C

Storage conditions: The kits are shipped at room temperature.

NOTE 1! Properly stored Kits are guaranteed until the expiry date. Note that shipping is realized at room temperature and will not affect stability. All components of the kit have been prepared under nucleases free conditions and have been thoroughly tested to ensure optimal performance.

NOTE 2! Storage conditions

All reagents in the kit can be stored at room temperature, except for the Proteinase K which has to be absolutely stored at +2-8°C. This will not affect the stability products.

For convenience, you can store the whole kit at +2-8°C. In this case, before using the kit, it is recommended to take out the reagents in advance and check if there are any precipitates. If the Buffers present precipitates place them at room temperature and eventually put them at +37°C.

IMPORTANT! Do not freeze the magnetic particles.

3. Equipment and reagents to be supplied by the user

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. To avoid contamination of your sample, wear face mask.

Reagents:

- 3M Dithiothreitol (DTT)
- Isopropanol
- 96-100% ethanol
- 70% ethanol

Materials:

- 56°C heat block, thermomixer or water bath
- Microtubes
- AutoMag Instrument (Cat.# 21101)
- Microtiter Deep well 96 plate (Cat.# 21102)
- 12-tip comb for 96 Deepwell plate (Cat.# 21103)
- Eution strip (Cat.# 21104)

Crime Prep Adem-Kit Protocol

The standard protocol is appropriate for all kind of sample types, such as samples on solid support or differential extraction samples. 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 purified using a Proteinase K treatment generally exhibits better locus-to-locus balance in downstream STR analysis.

1. Reagents preparation



WARNING ! CHEMICAL HAZARD. LB Buffer in contact with acids or bleach liberates toxic gases. Harmful if inhaled, absorbed through the skin, and swallowed. Causes eye, skin, and respiratory tract irritation. DO NOT ADD acids or bleach to any liquid wastes containing this product. Avoid breathing vapour. Do not taste or swallow. Use with adequate ventilation. Avoid contact with eyes and skin. Read the MSDS, and follow the handling instructions. Wear appropriate protective eyewear, clothing and gloves.



WARNING ! CHEMICAL HAZARD. Washing Buffer I Concentrate causes eye, skin and respiratory tract irritation. Avoid breathing vapour. Use with adequate ventilation. Avoid contact with eyes and skin. Read the MSDS, and follow the handling instructions; Wear appropriate protective, eyewear, clothing, and gloves.

- Prepare a **3M DTT** solution to perform lysis.

NOTE! You can prepare a 3M DTT stock solution and make aliquots for mid-term storage at -20°C.

- Prepare a 70% ethanol solution
- Prepare the Washing Buffers I & II before first use:
 1. Add 32mL of 96-100% ethanol to the bottle containing 24mL of Washing Buffer I. Homogenize the solution.
 2. Add 39mL of 96-100% ethanol to the bottle containing 17mL of Washing Buffer II. Homogenize the solution.

NOTE! Washing Buffer I & II are delivered concentrated. Before the first use, you have to **add ethanol** in the bottle in the indicated proportion.

2. Perform lysis

NOTE! Thaw an aliquot of frozen 3MDTT solution to prepare the lysis.

1. Place the sample at the bottom of a microtube.
2. Add **400µL of Crime Lysis Buffer** to the microtube containing the sample.
3. Add **50µL of Proteinase K** to the microtube containing the sample.
4. Add **4.5µL of 3M DTT** to the microtube containing the sample.

NOTE! It is recommended to prepare a fresh premixed lysis solution by combining 400µL of Crime Lysis Buffer, 50µL of Proteinase K and 4.5µL of 3M DTT per test.

Crime Lysis Buffer	400µL	} X number of extractions
Proteinase K	50µL	
3M DTT	4.5µL	

In this case, add 454.5µL of premixed lysis solution to the microtube containing the sample.

Then proceed to the incubation.

5. Close the tube and place it in a thermomixer, then incubate at +56°C and 1000rpm for 1 hour.

NOTE! For bone, tooth and adhesive samples, we recommend an incubation time at +56°C and 1000rpm for 2 hours.

IMPORTANT! You can use a heat block or a water bath instead of a thermomixer. For effective recovery, make sure that the sample is immersed by the Lysis Solution during incubation.

6. Perform DNA extraction with 400µL of Lysate

3. Instruction for AutoMag Instrument with 12-pin magnet head and 96 deep well plates for DNA extraction from forensic samples

These instructions are for the DNA extraction from 400µl of lysate using the Crime Prep Adem Kit (Cat.# 06210) and the AutoMag Instrument Duo (Cat.# 21101) with a 12-pin magnet head and 96 deep well plates.

AutoMag Instrument is a low-to-medium-throughput purification system with a magnetic particle processor for DNA purification kits. With the AutoMag Instrument customers can process up to 12 samples per run with a working volume up to 1 ml. In addition, it is possible to run two purification methods sequentially without interruption, raising the throughput up to 24 samples.



AutoMag Instrument (Cat.# 21101)

Instructions:

1. Prepare the lysate according to the instructions on page 8.
2. Take one empty Microtiter Deep well 96 plate and one Elution strip.



Microtiter Deep well 96 plate
(Cat.# 21102)



Elution strip
(Cat.# 21104)

3. Prepare the plate

Add the following reagents to the rows. **Note that row B is reserved for the tip comb and should be left empty.** Note that row A, G and H are left empty.

Plate name and type	Row	Row name	Content	Reagent/Sample volume per well
Microtiter Deep well 96 plate	A	Empty	Empty	Empty
	B	Tip	12-tip comb	Empty
	C	Sample	Lysate	400µL
			LB Buffer	250µL
			Isopropanol	250µL
			Silica-MasterBeads	15µL
	D	Wash 1	Washing Buffer I	500µL
	E	Wash 2	Washing Buffer II	500µL
	F	Wash 3	70% ethanol	500µL
	G	Empty	Empty	Empty
	H	Empty	Empty	Empty

NOTE! It is possible to prepare a premixed solution of Isopropanol / Silica-MasterBeads.

Isopropanol	250µL	]	X number of extractions
Silica-MasterBeads	15µL		

Fill up the **Row C** with **265µL of the premixed solution, plus 250µL of LB Buffer** and **400µL of Lysate**.



WARNING ! CHEMICAL HAZARD. LB Buffer in contact with acids or bleach liberates toxic gazes. Harmful if inhaled, absorbed through the skin, and swallowed. Cause eye, skin, and respiratory tract irritation. DO NOT ADD acids or bleach to any liquid wastes containing this product. Avoid breathing vapour. Do not taste or swallow. Use with adequate ventilation. Avoid contact with eyes and skin. Read the MSDS, and follow and handling instructions. Wear appropriate protecte eyewear, clothing and gloves.

4. Fill the Elution strip as follows.

Make sure that the elution strip is placed in the correct direction into the elution block. Ensure that the perforated end is facing towards the user and the Elution Buffer is pipetted into the correct wells.

Elution strip	Content	Reagent volume per well
Elution strip	Elution Buffer	60µL

5. Place a 12-tip comb for 96 Deepwell plate into row B of the plate.



12-tip comb for 96 Deewell plate (Cat.# 21103)

6. Start the Crime Prep protocol with the AutoMag Instrument and load the plate and elution strip.

*Switch ON the AutoMag Instrument and make sure that you are using the 12-pin magnet head and heating block. Start the Crime Prep protocol. Insert the plate and elution strip into the instrument as indicated on the AutoMag Instrument display and press **OK**. Make sure that the elution strip is placed in the correct direction into the elution block. Ensure that the perforated end is facing towards the user.*

7. After the run is completed, remove the plate and store the purified DNA.

When the protocol is completed, remove the plate and elution strip according to the instructions on the AutoMag Instrument display and turn off the instrument.

NOTE! Store or analyze the purified DNA accordingly. If DNA is not analyzed immediately, store it at 4°C for up to 24 hours. For longer period, consult laboratory guidelines. Freezing samples at -20°C has been shown to preserve DNA for longer periods of time.

Table 1 Summary of plate and elution strip content

Plate name and type	Row	Row name	Content	Reagent/Sample volume per well
Microtiter deep well 96 plate	A	Empty	Empty	Empty
	B	Tip	12-tip com	Empty
	C	Sample	Lysate	400µL
			LB Buffer	250µL
			Isopropanol	250µL
			Silica-MasterBeads	15µL
	D	Wash 1	Washing Buffer I	500µL
	E	Wash 2	Washing Buffer II	500µL
	F	Wash 3	70% ethanol	500µL
	G	Empty	Empty	Empty
	H	Empty	Empty	Empty
Elution strip		Elution	Elution Buffer	60µL

Troubleshooting

Observations	Comments and suggestions	
Error message in instrument display	Refer to the user manual supplied with your AutoMag Instrument.	
The tip comb holder lifting mechanism is out of position	Switch the instrument OFF and ON, and try again. If the error appears during initialization or is otherwise repeated, contact service.	
The turntable rotating mechanism is out of position	Switch the instrument OFF and ON, and try again. If the error appears during initialization or is otherwise repeated, contact service.	
The magnet head holder lifting mechanism is out of position	Switch the instrument OFF and ON, and try again. If the error appears during initialization or is otherwise repeated, contact service.	
The plastic tip comb is not attached to the holder	Check if the tips are presents. If it looks all right, turn ON and OFF, and run the check protocol.	
Nor or low yield of DNA	Biological sample contains no or low amount of DNA.	Review protocol steps and reagents additions.
		Extract DNA from a different cutting from the sample

Warranty

This product is only for use in research. The purchaser is responsible to validate the performance of this product for any particular use, and to use the product in compliance with any applicable regulations.

The products are warranted to the original purchaser only to conform to the quality and contents stated on the vial and outer labels for duration of the stated shelf life. Ademtech's obligation and the purchaser's exclusive remedy under this warranty is limited either to replacement, at Ademtech's expense, of any products which shall be defective in manufacture, and which shall be returned to Ademtech, transportation prepaid, or at Ademtech's option, refund of the purchase price. Claims for merchandise damaged in transit must be submitted to the carrier.

Ordering Information

- **Ademtech Kits**

CAT NO.	PRODUCT	PACKAGE SIZE
06210	Crime Prep Adem-Kit	1 x 96
06140	Smart D-N-Adem-Kit for Profiling	1 x 100

- **Instrument and consumables**

CAT NO.	PRODUCT	PACKAGE SIZE
21101	AutoMag Instrument	Each
21102	Microtiter Deepwell 96 plate	50 pcs
21103	12-tip comb for 96 Deepwell plate	50 pcs
21104	Elution strip	40 pcs
21105	Combi pack for 96 Deepwell plate	Each