



Ron's Gel Extraction Mini Kit

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Kit for extraction of DNA fragments from standard or low-melt agarose gels

Research Use Only (RUO)

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<b>Ron's Gel Mini Extraction Kit</b>	Ref. No: 802501 (50 preps)
	Ref. No: 802501L (5 x 50 preps)
Valid from:	August 2019



## Ron's Gel Extraction Mini Kit

### 1. Introduction

**Ron's Gel Extraction Kits** provide an easy, safe and reliable method for extraction of DNA fragments from standard or low-melt agarose gels in TAE or TBE buffer. The procedure is based on optimized buffers and the use of our specially designed **Ron's Spin Columns** with a high binding capacity. The advanced buffer system is optimized for efficient recovery of DNA and removal of contaminants. DNA is adsorbed to the Ron's Spin Membrane and all impurities are efficiently removed by washing and centrifugation. The pure DNA is directly eluted in a special buffer.

The components of this kit are sufficient for processing samples of up to 100 mg - 150 mg agarose gel.

### 2. Content of the Kit

Ref No	S802501 10 preps (sample size)	802501 50 preps	802501L 5 x 50 preps
Mini spin columns	10	50	5 x 50
Collection tubes 2.0 ml	10	50	5 x 50
Lysis Buffer LG-1	25 ml	25 ml	5 x 25 ml
Wash Buffer concentrate WB-2 *	5 ml	2x 5 ml	10 x 5 ml
Elution Buffer EL-3	6 ml	6 ml	5 x 6 ml
Manual	1	1	1

\* Add ethanol

### 3. Storage Conditions and Stability

All components of the **Ron's Gel Extraction Kit** should be stored dry at room temperature (15 - 25 °C). Under these conditions the kit can be stored for 24 months. Guarantee for full performance of the kit as specified in this manual is only valid if storage conditions are followed.

### 4. Quality Control

The performance of the **Ron's Gel Extraction Kit** is monitored routinely on a lot-to-lot basis.



## Ron's Gel Extraction Mini Kit

### 5. Safety Information

The following components of **Ron's Gel Extraction Kit** contain hazardous contents. It is strongly recommended to wear a lab coat, disposable gloves and protective goggles when working with chemicals. More detailed information is available in the material safety data sheets, which can be requested from the manufacturer. There is no need of labeling harmful features with H & P phrases upon packing sizes of 125 ml or 125 g.

**Caution:** Do not add bleach or acidic solutions to the waste of sample preparation!

Component	Hazard content	GHS symbol		Hazard phrases	Precaution phrases
Lysis Buffer LG-1	Guanidine hydrochloride 36-50%		Warning	302, 319	280, 301+312 305+351+338, 330, 337+313

Hazard phrases	
H302	Harmful if swallowed
H319	Causes serious eye irritation

Precaution phrases	
P280	Wear protective gloves / eye protection
P301+312	If swallowed: call a poison center/doctor/ .../ if you well unwell
P305+351+338	If in eyes: rinse cautiously with clean water for several minutes. Remove contact lenses. Continue rinsing
P330	Rinse mouth
P337+313	If eye irritation persists: get medical advice/attention

### 6. Protocol

#### Additional Material Required:

- 96-100 % ethanol
- 100 % isopropanol
- Incubator/ heat shaker or water bath
- Microcentrifuge
- Receiver tubes (1.5 ml)

#### Before starting:

Wash Buffer WB-2 is a concentrate. Before using for the first time, add the appropriate amount of ethanol (96 - 100 %) as indicated on the bottle and in the table below:

Kit size	Wash Buffer WB-2	Ethanol to be added	Final volume
10, 50	5 ml	21 ml	26 ml

## **Ron's Gel Extraction Mini Kit**

### **Protocol**

This protocol is designed for the extraction and purification of DNA from up to 100 -150 mg standard or low-melt agarose gel per spin column.

1. Excise the DNA fragment from the agarose gel with a sterilised tip or scalpel.
2. Weigh the gel slice in a colourless tube (1.5 ml tube). **Add 3 volumes of Lysis Buffer LG-1 to 1 volume of gel.** (Example: For 100 mg gel use 300 µl Lysis Buffer LG-1).
3. Incubate at 50 °C for 10 min (or until the gel slice has completely dissolved). To help dissolve gel, mix by vortexing the tube every 2 - 3 minutes during incubation or use a heat shaker.
4. **Add 1 gel volume isopropanol to the sample** and mix by vortexing. (Example: for **100 mg gel add 100 µl isopropanol**).
5. Place a spin column in a provided 2 ml collection tube.
6. To **bind DNA**, apply the sample to the column and centrifuge for 30 - 60 s at 10000 g (approx. 13000 rpm).
7. Discard flow-through. Place the column back into the same tube. Collection tubes are re-used to reduce plastic waste.
8. **For washing** add 500 µl **Wash Buffer WB-2** (add ethanol before use, as indicated on the bottle) to the column and centrifuge for 30 - 60 s at 10000 g (approx. 13000 rpm).
9. Discard flow-through. Place the column back into the same tube.
10. **Second wash step**: add 500 µl **Wash Buffer WB-2** to the column and centrifuge for 2 min at 10000 g.
11. Discard flow-through.
12. Place column in a clean 1.5 ml receiver tube (not included) and heat the column at 70°C for 5 minutes to remove rest of alcohols.
13. **Elute DNA**: add 30 µl - 50 µl **Elution Buffer EL-3** (pre-heated to 70°C) to centre of the column membrane, incubate at room temperature (18 - 25°C) **for 1 minute** and centrifuge the column for 1 min at 10000 g to collect the eluate.

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### 7. Trouble Shooting

This guide can help solve problems that may arise.

Observation	Possible cause	Suggestions
Clogged column	Incomplete lysis	If using more than 100 mg of agarose gel sample, increase volumes of buffers /ethanol in proportion. Avoid overloading the spin columns. Mix by vortexing every 2-3 minutes during incubation to ensure complete lysis of the gel slice.
Poor or low recovery	Improper washing Poor elution	Confirm the wash solution concentrates were diluted with the specified volume of ethanol. Repeat elution or increase elution volume. High recovery: from $\leq 100$ mg gel slice per spin column.
Low A260/280 ratio	Purification is incomplete due to column overloading or inadequate lysis	Reduce sample volume. Low yields and impure DNA are attributable if the system is overloaded. Ensure the gel slice has been completely solubilized
Low DNA performance	Salt in eluate	Make sure that you followed all washing steps of the procedure.

### 8. Warranty and Guarantee of Products

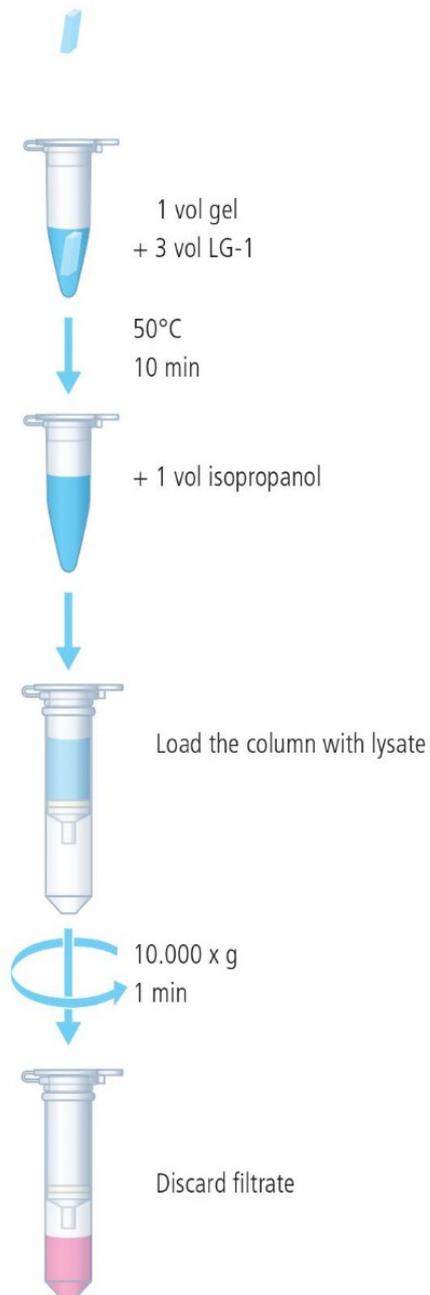
The manufacturer guarantees the performance of its Ron's Gel Extraction Kit in the manner described in this handbook. It is up to the user to determine the suitability of Ron's Gel Extraction Kit for its particular use. In case a product fails to perform due to any reason except misuse, the manufacturer will replace it without further charge or refund the purchase price. We reserve the right to change, alter, or modify our Ron's Gel Extraction Kit to enhance its performance and design. The manufacturer's terms and conditions are available on request.

### 9. Limitations of Product Use

The use of all products of **Ron's Purification Kits** is strictly limited to research purposes. They are not to be applied for any diagnostic use, including human or drug purposes.

## Ron's Gel Extraction Mini Kit

### 10. Flowchart of Extraction



**Step 1: Excision of DNA fragment**

**Step 2: Lysis / solubilisation**

**Step 3: Binding DNA on column**

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