

## Manual

# **ExToPCR™**

Fast, enzymatic extraction of DNA from various samples.
The DNA extract can be used for standard PCR or real-time PCR.

catalog#	size
1032-100	100 reactions
1032-500	500 reactions

For research use only.

#### Guarantee

A&A Biotechnology provides guarantee on this product.

The company does not guarantee correct performance of this kit in the event of:

- not adhering to the supplied protocol
- use of not recommended equipment or materials
- use of other reagents than recommended or which are not a component of the product
- use of expired or improperly stored product or its components

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## **Advantages**

- Fast, 15 minute procedure.
- DNA extraction is performed in a single-tube, without the need for multiple washing steps or centrifugation.

## **Specification**

form	buffer-based DNA extraction	
sample type	<ul> <li>blood</li> <li>FFPE tissue</li> <li>swabs</li> <li>hair follicle</li> <li>animal tissue</li> <li>insect</li> <li>feathers</li> </ul>	

## **Description**

Fast lysis procedure allows for efficient extraction of DNA in an amount required for PCR or real-time PCR reactions. Thermostable **XTP enzyme** and dedicated **XTP buffer** ensures optimal extraction efficiency and inactivation of cellular nucleases. In addition, the extraction buffer does not contain harmful and irritating substances.

DNA extracted with ExToPCR™ can be used with any standard PCR reagents. However, for best results we recommend following PCR mixes:

#### Standard PCR

- PCR Mix Plus Green 2005-100Z, 2005-1000Z
- PCR Mix Plus Red 2005-100P, 2005-1000P
- PCR Mix Plus Clear 2005-100C, 2005-1000C
- PCR Mix Plus HGC 2005-100G, 2005-1000G

#### Real-time PCR

- <u>RT PCR Mix EvaGreen</u> 2008-100G, 2008-1000G
- RT PCR Mix Probe 2008-200P, 2008-2000P
- RT PCR Mix Sybr 2008-100, 2008-1000

#### Hot Start real-time PCR

- RT HS-PCR Mix Probe 2017-200P, 2017-2000P
- RT HS-PCR Mix Sybr 2017-100HS, 2017-1000HS
- qPCR-HS Mix EvaGreen 2008HS-100G, 2008HS-1000G
- qPCR-HS Mix Probe 2008HS-100P, 2008HS-1000P
- qPCR-HS Mix Sybr 2008HS-100, 2008HS-1000

### **Contents**

component	1032-100	1032-500	storage
XTP buffer	10 ml	5 x 10 ml	-20 °C
XTP enzyme	500 µl	2 x 1.3 ml	-20 °C

# Additional equipment and reagents

### **Necessary**

- 0.2 ml PCR tubes
- Thermoblock, thermocycler or water bath

## **Optional**

- TE buffer or Tris-HCl pH 8.0
- 1.5 ml Eppendorf tubes

## **Sample preparation**

sample type	preparation	
	1. Add to 0.2 ml tube:	
Blood	o <b>5-10 μl</b> of fresh or EDTA blood	
	o 85 μl of XTP buffer	
	o 5 μl of XTP enzyme	
	2. Follow the extraction protocol.	
FFPE tissue	Trim all excess wax from FFPE tissue sample	
	2. Add to 0.2 ml tube:	
	$\circ~~$ 1 mm³ or 1–2 mm² fragment of 10 $\mu$ m section FFPE tissue sample	
	o 85 μl of XTP buffer	
	o 5 μl of XTP enzyme	
	3. Follow the extraction protocol.	
Swabs	1. Place cut off swab in 1.5 ml tube and add:	
	o 300 μl of 0.5X XTP buffer (diluted in water)	
	o 5 μl of XTP enzyme	
	2. Follow the extraction protocol.	

	1.	Add to 0.2 ml tube:	
		o <b>1-10</b> individual follicles	
Hair follicle		o 85 μl of XTP buffer	
		o 5 μl of XTP enzyme	
	2.	Follow the extraction protocol.	
	1.	Add to 0.2 ml tube:	
		o 2 mm³ tissue fragment	
Animal tissue		o 85 μl of XTP buffer	
		o 5 μl of XTP enzyme	
	2.	Follow the extraction protocol.	
	1.	Place the insect in a 1.5 ml tube	
	2.	Add XTP buffer to immerse the entire insect	
Insects	3.	Crush insect with a pipette tip or other sterile tool	
	4.	Add 5 µl of XTP enzyme	
	5.	Follow the extraction protocol.	
	1.	Place <b>2-5 mm</b> quill fragment in a 0.2 ml tube and add:	
		o 100 μl of XTP buffer	
Feathers		o 20 μl of XTP enzyme	
	2.	Follow the extraction protocol.	

# **Extraction protocol**

- 1. Close the tube with the sample.
- 2. Incubate in a waterbath, thermoblock or thermocycler for 10 min at 50 °C.
- 3. Incubate in a waterbath, thermoblock or thermocycler for 5 min at 95 °C.
- 4. Keep the sample at **room temp.** to cool down.

**Note**: If the sample fragment does not dissolve completely do not remove it from the tube. Despite the presence of a sample fragment, the DNA present in the extract is safe.

5. Follow up with PCR or store DNA extract at 4 °C for up to 1 month.

# **Troubleshooting**

## Thick lysate after DNA extraction

If the lysate is thick or there is a problem with pipetting centrifuge it briefly and use supernatant in PCR. Alternatively dilute lysate 1:5 - 10 with TE buffer or Tris-HCl pH 8.0.

### **PCR** inhibition

In case of non-specific or lack of PCR product, dilute the lysate 1:5 – 10 with TE buffer or Tris-HCl pH 8.0.

## **Safety information**

#### XTP enzyme



H315 Causes skin irritation.

H319 Causes serious eye irritation.

H334 May cause allergy or asthma symptoms or breathing difficulties if inhaled.

H335 May cause respiratory irritation.

P261 Avoid breathing dust.

P305+P351+P338 If in eyes: rinse cautiously with water for several minutes.

Remove contact lenses,

if present and easy to do. Continue rinsing.

 ${\sf P342+P311\ If\ experiencing\ respiratory\ symptoms\ call\ a\ Poison\ Center\ or\ doctor\ /}$ 

physician.



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