

# Produktinformation



Forschungsprodukte & Biochemikalien



Zellkultur & Verbrauchsmaterial



Diagnostik & molekulare Diagnostik



Laborgeräte & Service

Weitere Information auf den folgenden Seiten! See the following pages for more information!



### Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

## Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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### Quick-DNA™ 96 Kit

High throughput isolation of ultra-pure DNA from biological liquids and cells.

#### **Highlights**

- Quick high throughput (96-well) purification of high-quality DNA from whole blood, plasma, serum, bodily fluids, buffy coat, lymphocytes, swabs or cultured cells in less than 25 minutes using innovative Zymo-Spin<sup>™</sup> Technology.
- Compatible with commonly used anticoagulants (i.e., EDTA, heparin, citrate).
- Unique extraction technology excludes the use of Proteinase K and organic denaturants.
- Isolated DNA is ideal for PCR, endonuclease digestion, bisulfite conversion/methylation detection, sequencing, genotyping, etc.

Catalog Numbers: D3010. D3011. D3012



Scan with your smart-phone camera to view the online protocol/video.







# **Table of Contents**

Product Contents	01
Specifications	02
Product Description	03
Buffer Preparation	04
Protocols	04
Whole Blood, Serum, and Plasma Samples	. 04
Buccal Cells and Swabs	05
Solid Tissue Samples	06
Cell Monolayer Samples	07
Cell Suspensions and Proteinase K	
Digested Samples	08
Samples in DNA/RNA Shield <sup>™</sup>	. 09
Proteinase K Digestion with	
DNA/RNA Shield™	10
Troubleshooting	11
Ordering Information	12
Complete Your Workflow	13
Guarantee	17

Revised on: 7/5/2022

## **Product Contents**

<i>Quick</i> -DNA <sup>™</sup> 96 Kit	<b>D3010</b> (2 x 96 Preps)	<b>D3011</b> (4 x 96 Preps)	<b>D3012</b> (10 x 96 Preps)	Storage Temperature
Genomic Lysis Buffer <sup>1</sup>	100 ml	100 ml (2x)	100 ml (5x)	Room Temp.
DNA Pre-Wash Buffer <sup>2</sup>	50 ml	50 ml (2x)	50 ml (5x)	Room Temp.
g-DNA Wash Buffer	100 ml	100 ml (2x)	100 ml (5x)	Room Temp.
DNA Elution Buffer	10 ml	10 ml (2x)	50 ml	Room Temp.
Silicon-A™ Plate	2	4	10	Room Temp.
Collection Plate	2	4	10	Room Temp.
Elution Plate	2	4	10	Room Temp.
Instruction Manual	1	1	1	-

<sup>&</sup>lt;sup>1</sup> <u>Recommended</u>: Add beta-mercaptoethanol to 0.5%(v/v) i.e., 250 μl per 50 ml *or* 500 μl per 100 ml prior to use.
<sup>2</sup> A precipitate may have formed in the DNA Pre-Wash Buffer during shipping. To completely resuspend the buffer, incubate the bottle at 30-37 °C for 30 minutes and mix by inversion. DO NOT MICROWAVE.

## **Specifications**

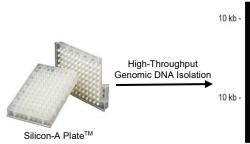
- Sample Sources<sup>1</sup> Whole blood, plasma, or serum from humans, mice, rats, etc. Also, cells from culture, buccal cells, as well as a variety of biological liquids are effectively processed using this kit. Tissue already digested with Proteinase K or mechanically homogenized can also be processed.
- Workflow Overview Unique lysis buffer system omits the need for Proteinase K digestion for biological fluids and cell culture samples.
- DNA Purity High-quality DNA is eluted with DNA Elution Buffer or water. DNA is especially well suited for PCR and other downstream applications. A<sub>260</sub>/A<sub>280</sub>>1.8
- DNA Size Limits Capable of recovering genomic DNA up to and above 40 kb. In most instances, mitochondrial DNA and viral DNA (if present) will also be recovered.
- DNA Recovery Up to 5 µg/well of total DNA is eluted into ≥ 30 µl DNA Elution Buffer or water. Human whole blood will typically yield 1.5-3.5 µg DNA per 50 µl blood sampled. Mammalian tissues already homogenized yield: 1-3 µg DNA per mg skeletal, heart, and brain tissues and 3-5 µg DNA per mg liver, kidney, and lung tissues.
- **Product Detergent Tolerance** ≤5% Triton X-100, ≤5% Tween-20, ≤5% Sarkosyl, ≤0.1% SDS
- Equipment Microcentrifuge, vortex, centrifuge with microplate carriers

<sup>&</sup>lt;sup>1</sup>For high-throughput DNA purification from biological fluids, cell cultures, and solid tissues utilizing Proteinase K, use the *Quick*-DNA™ 96 Plus Kit (D4070, D4071).

## **Product Description**

The *Quick-DNA™* **96 Kit** is a simple, high throughput (96-well) procedure for the rapid isolation of total DNA (e.g., genomic, mitochondrial, viral) from a variety of biological sample sources. This product has been optimized for maximal recovery of ultra-pure DNA without RNA contamination and is compatible with whole blood (fresh or stored), serum, plasma, buffy coat, buccal cells, cells from culture, and many biological liquid samples.

For processing, simply add the specially formulated **Genomic Lysis Buffer** to a sample, vortex, and transfer the mixture to the supplied **Silicon-A™ Plate**. There is no need for organic denaturants or Proteinase K digestion because of the unique lysis buffer system. The product features **Zymo-Spin™ Technology** to yield high-quality, purified DNA in just minutes (see below). PCR inhibitors are effectively removed during the purification process. DNA purified using the **Quick-DNA™ 96 Kit** is suitable for PCR, nucleotide blotting, DNA sequencing, restriction endonuclease digestion, bisulfite conversion/methylation analysis, and other downstream applications.



M 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15

M 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30

Genomic DNA isolated from mouse tailsnips using the *Quick*-DNA™ 96 Kit. A total of 30 mouse tailsnips were homogenized with Zymo Research's Squisher-8™ then processed using the *Quick*-DNA™ 96. About one third of the number of eluted DNAs was then separated in a 0.8% w/v agarose gel (shown in lanes 1 to 30).

#### **Protocol**

#### **Buffer Preparation:**

✓ <u>Recommended</u>: Add beta-mercaptoethanol (user supplied) to the **Genomic Lysis Buffer**<sup>1</sup> to a final dilution of 0.5%(v/v) i.e., 250 µl per 50 ml or 500 µl per 100 ml.

#### Whole Blood, Serum, and Plasma Samples

The following is for the purification of DNA from 50  $\mu$ l whole blood, serum or plasma (the volumes can be adjusted up to 100  $\mu$ l (max.) depending on your requirements). Fresh, frozen, or preserved blood (in EDTA, citrate, or heparin) can be used. If material cannot be processed immediately, the sample can be "stabilized" for later processing (see page 5) although the immediate processing of blood samples is recommended.

- 1. Add 200 µl of **Genomic Lysis Buffer** to 50 µl of blood, serum<sup>2</sup>, or plasma (4:1) <sup>3,4</sup>. Mix completely by vortexing 4-6 seconds, then let stand 5-10 minutes at room temperature.
- Transfer the mixture to the wells of a Silicon-A™ Plate<sup>5</sup> on a Collection Plate. Centrifuge at 2,500 - 5,000 x g for 5 minutes.
- 3. Add 200 μl of **DNA Pre-Wash Buffer** to each well and centrifuge at 2,500 5,000 *x g* for 5 minutes. Discard the flow through.
- Add 300 µl of g-DNA Wash Buffer to each well and centrifuge at 2,500 -5,000 x g for 5 minutes.
- 5. Transfer the Silicon-A<sup>TM</sup> Plate onto an Elution Plate. Add  $\geq$  30  $\mu$ l DNA Elution Buffer or water<sup>6,7</sup> to each well. Incubate 2-5 minutes at room temperature and then centrifuge at 2,500 5,000  $\times$   $\times$   $\times$  for 5 minutes to elute the DNA. The eluted DNA can be used immediately for molecular based applications or stored  $\leq$  -20°C for future use.

<sup>&</sup>lt;sup>1</sup> The addition of BME to the Genomic Lysis Buffer is recommended for optimal performance when working with protein rich samples (tissue, whole blood, etc.). If BME is not added, please ensure the sample is thoroughly homogenized in the Genomic Lysis Buffer prior to purification.

<sup>&</sup>lt;sup>2</sup> For the inclusion of <u>small DNAs</u> from serum, add 0.3 volumes isopropanol to the mixture. (For example, to a 1 ml mixture of serum and Genomic Lysis Buffer add 300 µl isopropanol.)

<sup>&</sup>lt;sup>3</sup>Add 200 µl Genomic Lysis Buffer to all samples <50 µl.

<sup>&</sup>lt;sup>4</sup> Step 1 can be performed directly in the Silicon-A™ Plate. Mixing is performed by pipetting the samples up and down repeatedly.

<sup>&</sup>lt;sup>5</sup>The capacity of each well of the Silicon-A™ Plate is ~600 μl.

<sup>&</sup>lt;sup>6</sup> Elution of DNA from the column is dependent on pH and temperature. If water is used, ensure the pH is >6.0. Also, the total yield may be improved by eluting the DNA with Elution Buffer or water pre-equilibrated to 60-70°C. 
<sup>7</sup> The **DNA Elution Buffer** contains 10 mM Tris-HCl, pH 8.5, 0.1 mM EDTA. If required, pure water can also be

used to elute the DNA.

<u>Delayed Processing (Stabilization) of Blood Samples:</u> The immediate processing of blood with this kit is recommended. However, if blood cannot be processed immediately, samples can be "stabilized" in **Genomic Lysis Buffer** for processing at a later time. To do this, add *four* volumes of Genomic Lysis Buffer to *each* volume of whole blood (4:1), then vortex. Blood samples mixed with Genomic Lysis Buffer can be stored at room temperature for 1-2 weeks, 0-4 °C for 1-2 months, -20 °C for 6 months to a year, or <-70 °C for many years. Samples stored at ≤ 4 °C should reach room temperature prior to processing. Begin at Step 2 in the standard protocol on page 4 when purifying DNA from blood samples stabilized in Genomic Lysis Buffer.

#### **Buccal Cells and Swabs**

Buccal cells can be isolated using a rinse- or swab-based isolation method.

- A. **Rinse Method**: Vigorously rinse 10-20 ml of saline solution or mouthwash orally for 30 seconds. The more vigorous the rinsing action, the more cells that will be recovered. Spit the saline into a 50 ml tube and pellet the cells at 1,500 rpm for 5 minutes. Discard the supernatant without disturbing the cell pellet. Add 500 µl of **Genomic Lysis Buffer** to the pellet then vortex 4-6 seconds, then let stand at room temperature for 5-10 minutes.
- B. **Swab Isolation Method**: Thoroughly rinse mouth out before isolating cells. Brush the inside of the cheek with a *buccal swab* for 15 seconds (approximately 20 brushes), making sure to cover the entire area of the inner cheek. Rinse the brush into a microcentrifuge tube using 500 µl of **Genomic Lysis Buffer**, vortex 4-6 seconds, and then let stand at room temperature for 5-10 minutes.
- Transfer the mixture to the wells of a Silicon-A™ Plate¹ on a Collection Plate. Centrifuge at 2,500 - 5,000 x g for 5 minutes.
- 2. Add 200  $\mu$ l of **DNA Pre-Wash Buffer** to each well and centrifuge at 2,500 5,000 x g for 5 minutes. Discard the flow through.
- 3. Add 300  $\mu$ l of **g-DNA Wash Buffer** to each well and centrifuge at 2,500 5,000 x g for 5 minutes.
- 4. Transfer the Silicon-A<sup>TM</sup> Plate onto an Elution Plate. Add  $\geq$  30 µl DNA Elution Buffer or water<sup>2,3</sup> to each well. Incubate 2-5 minutes at room temperature and then centrifuge at 2,500 5,000 x g for 5 minutes to elute the DNA. The eluted DNA can be used immediately for molecular based applications or stored  $\leq$  -20°C for future use.

<sup>&</sup>lt;sup>1</sup>The capacity of each well of the Silicon-A™ Plate is ~600 µl.

<sup>&</sup>lt;sup>2</sup> Elution of DNA from the column is dependent on pH and temperature. If water is used, ensure the pH is >6.0. Also, the total yield may be improved by eluting the DNA with Elution Buffer or water pre-equilibrated to 60-70°C.

<sup>&</sup>lt;sup>3</sup> The **DNA Elution Buffer** contains 10 mM Tris-HCl, pH 8.5, 0.1 mM EDTA. If required, pure water can also be used to elute the DNA.

#### Solid Tissue Samples

For Proteinase K digested materials (e.g., tailsnips) follow the protocol for **Cell Suspensions and Proteinase K Digested Samples** (see page 8). Otherwise, mechanically homogenize<sup>1</sup> <u>up to</u> 5 mg of fresh or frozen tissue in 500 µl of **Genomic Lysis Buffer.** 

- Centrifuge the lysate at ≥ 10,000 x g for 5 minutes. Making sure not to disturb the pelleted debris, transfer the supernatant to the wells of a Silicon-A<sup>™</sup> Plate² on a Collection Plate. Centrifuge at 2,500 5,000 x g for 5 minutes.
- 2. Add 200 μl of **DNA Pre-Wash Buffer** to each well and centrifuge at 2,500 5,000 *x g* for 5 minutes. Discard the flow through.
- Add 300 μl of g-DNA Wash Buffer to each well and centrifuge at 2,500 -5,000 x g for 5 minutes.
- 4. Transfer the **Silicon-A<sup>TM</sup> Plate** onto an **Elution Plate**. Add  $\geq$  30  $\mu$ l **DNA Elution Buffer** or water<sup>3,4</sup> to each well. Incubate 2-5 minutes at room temperature and then centrifuge at 2,500 5,000  $\times$   $\times$   $\times$  for 5 minutes to elute the DNA. The eluted DNA can be used immediately for molecular based applications or stored  $\leq$  -20°C for future use.

<sup>&</sup>lt;sup>1</sup>For solid tissues, Proteinase K treatment or mechanical homogenization is required. For high-throughput DNA purification from biological fluids, cell cultures, and solid tissues utilizing Proteinase K use the *Quick*-DNA™ 96 Plus Kit (D4070, D4071).

<sup>&</sup>lt;sup>2</sup>The capacity of each well of the Silicon-A™ Plate is ~600 µl.

<sup>&</sup>lt;sup>3</sup> Elution of DNA from the column is dependent on pH and temperature. If water is used, ensure the pH is >6.0.
Also, the total yield may be emproved by eluting the DNA with Elution Buffer or water pre-equilibrated to 60-70°C.

<sup>&</sup>lt;sup>4</sup>The **DNA Elution Buffer** contains 10 mM Tris-HCl, pH 8.5, 0.1 mM EDTA. If required, pure water can also be used to elute the DNA.

#### Cell Monolayer Samples

The following procedure is designed for <u>up to</u> 1.0x10<sup>6</sup> (max.) monolayer cells<sup>1</sup> (roughly equal to one well of a 6-well plate). Although cell types and culture conditions may vary, the protocol will work with high-density growth cells (e.g., HeLa cells) as well as with low-density growth cells (e.g., neuronal cells). The procedure may be scaled up or down for increases or decreases in the amounts of monolayer cells sampled (see the **Guidelines for Monolayer Cell DNA Isolation** below).

- 1. Trypsinize or manually scrape adherent cells² from the growth surface of a culture flask or plate. Centrifuge the cell suspension at approximately 500 *x g* for 5 minutes. Remove the supernatant and add 500 µl of **Genomic Lysis Buffer** directly to the cell pellet. Resuspend pellet by vortexing 4-6 seconds and let stand for 5-10 minutes at room temperature.
- 2. Transfer the mixture to the wells of a Silicon-A<sup>TM</sup> Plate<sup>3,4</sup> on a Collection Plate. Centrifuge at  $2,500 5,000 \times g$  for 5 minutes.
- 3. Add 200 μl of **DNA Pre-Wash Buffer** to each well and centrifuge at 2,500 5,000 *x g* for 5 minutes. Discard the flow through.
- Add 300 µl of g-DNA Wash Buffer to each well and centrifuge at 2,500 -5,000 x g for 5 minutes.
- 5. Transfer the Silicon-A<sup>TM</sup> Plate onto an Elution Plate. Add  $\geq 30 \mu l$  DNA Elution Buffer or water<sup>5,6</sup> to each well. Incubate 2-5 minutes at room temperature and then centrifuge at 2,500 5,000 x g for 5 minutes to elute the DNA. The eluted DNA can be used immediately for molecular based applications or stored  $\leq$  -20°C for future use.

<u>Guidelines for Monolayer Cell DNA Isolation:</u> The above procedure is designed for the processing of  $0.1-1.0x10^6$  cells. However, cell numbers (growth densities) can vary between different cell types. Table 1 on page 8 provides an approximation of what can be recovered from different culture containers for high-density growth cells like CV1 and HeLa cells. If processing more than  $1.0x10^5$  cells, <u>double</u> the volume of **Genomic Lysis Buffer** added (i.e., 1000 μl) to the sample.

<sup>&</sup>lt;sup>1</sup>Generally, no more than 1.0x10<sup>6</sup> cells should be sampled; larger samples will exceed the binding capacity of the spin column. See <u>Guidelines for Monolayer Cell Isolation</u> (see above).

<sup>&</sup>lt;sup>2</sup>Alternatively: Cells can be lysed directly in the culture container by removing the medium and adding the Genomic Lysis Buffer directly to the monolayer surface.

<sup>&</sup>lt;sup>3</sup> It may be necessary to centrifuge the sample mixture before transferring the supernatant to the Silicon-A™ Plate to remove insoluble material that may clog the column.

<sup>&</sup>lt;sup>4</sup>The capacity of each well of the Silicon-A<sup>™</sup> Plate is ~600 μl.

<sup>&</sup>lt;sup>5</sup> Elution of DNA from the column is dependent on pH and temperature. If water is used, ensure the pH is >6.0. Also, the total yield may be improved by eluting the DNA with Elution Buffer or water pre-equilibrated to 60-70°C.

<sup>&</sup>lt;sup>6</sup> The **DNA Elution Buffer** contains 10 mM Tris-HCl, pH 8.5, 0.1 mM EDTA. If required, pure water can also be used to elute the DNA.

Table 1: Culture Plate/Flask Growth Area (cm<sup>2</sup>) and Cell Number

Culture Container	Well /Flask Surface Area	Cell Number
96-well plate (each well)	$0.32 \text{-} 0.6 \text{ cm}^2$	4-5x10 <sup>4</sup>
24-well plate (each well)	2 cm <sup>2</sup>	1-3x10⁵
12-well plate (each well)	4 cm <sup>2</sup>	4-5x10⁵
6-well plate (each well)	9.5 cm <sup>2</sup>	0.5-1x10 <sup>6</sup>
T25 Culture Flask	25 cm <sup>2</sup>	2-3x10 <sup>6</sup>
T75 Culture Flask	75 cm <sup>2</sup>	$0.6-1x10^7$
T175 Culture Flask	175 cm <sup>2</sup>	2-3x10 <sup>7</sup>

#### Cell Suspensions and Proteinase K Digested Samples

The following protocol is designed for <u>up to</u> 100 µl of biological liquid sample including CSF, buffy coat, body fluids (semen), and cell suspensions¹ containing less than 1.0x10<sup>6</sup> cells as well as lysates derived from Proteinase K digested samples².

- Add 4 volumes of Genomic Lysis Buffer to each volume of liquid sample (4:1). (e.g., add 400 μl of Genomic Lysis Buffer to 100 μl liquid sample). Mix briefly by vortexing, then let stand at room temperature for 5-10 minutes<sup>3</sup>.
- Transfer the mixture to the wells of a Silicon-A™ Plate<sup>4</sup> on a Collection Plate. Centrifuge at 2,500 - 5,000 x g for 5 minutes.
- 3. Add 200 μl of **DNA Pre-Wash Buffer** to each well and centrifuge at 2,500 5,000 *x g* for 5 minutes. Discard the flow through.
- 4. Add 300 µl of **g-DNA Wash Buffer** to each well and centrifuge at 2,500 5,000 *x g* for 5 minutes.
- 5. Transfer the Silicon-A<sup>TM</sup> Plate onto an Elution Plate. Add  $\geq$  30  $\mu$ l DNA Elution Buffer or water<sup>5,6</sup> to each well. Incubate 2-5 minutes at room temperature and then centrifuge at 2,500 5,000 x g for 5 minutes to elute the DNA. The eluted DNA can be used immediately for molecular based applications or stored  $\leq$  -20°C for future use.

<sup>&</sup>lt;sup>1</sup>Cells should be processed directly from biological fluids or from suspension in PBS, TE, or compatible buffers.

<sup>&</sup>lt;sup>2</sup> For solid tissues, Proteinase K treatment or mechanical homogenization is required. For high-throughput DNA purification from biological fluids, cell cultures, and solid tissues utilizing Proteinase K use the *Quick*-DNA™ 96 Plus Kit (D4070, D4071).

<sup>&</sup>lt;sup>3</sup> For Proteinase K digested material, centrifuge the mixture at 10,000 *x g* for 5 minutes after incubating. Transfer up to 600 μl of supernatant to the wells of a Silicon-A<sup>TM</sup> Plate in Step 2.

<sup>&</sup>lt;sup>4</sup> The capacity of each well of the Silicon-A™ Plate is ~600 µl.

<sup>&</sup>lt;sup>5</sup>Elution of DNA from the column is dependent on pH and temperature. If water is used, ensure the pH is >6.0. Also, the total yield may be improved by eluting the DNA with Elution Buffer or water pre-equilibrated to 60-70°C

<sup>&</sup>lt;sup>6</sup> The DNA Elution Buffer contains 10 mM Tris-HCl, pH 8.5, 0.1 mM EDTA. If required, pure water can also be used to elute the DNA.

# Samples in DNA/RNA Shield<sup>™</sup>

**DNA/RNA Shield™** ensures nucleic acid stability during sample storage/transport at ambient temperatures. There is no need for refrigeration or specialized equipment. DNA/RNA Shield™ effectively lyses cells and inactivates nucleases and infectious agents (virus), and it is compatible with various collection and storage devices (vacutainers, swabs, nasal, buccal, fecal, etc.).

DNA/RNA Shield™ can be purchased separately (R1100-50 or R1100-250).

See page 10 for performing a Proteinase K Digestion on tissue samples stored in DNA/RNA Shield™.

- Add 400 µl of Genomic Lysis Buffer to 100 µl of the sample/shield mixture prepared according to the DNA/RNA Shield™ specifications (4:1).
- 2. Mix completely by vortexing 4-6 seconds, then let stand 5-10 minutes at room temperature<sup>1</sup>.
- 3. Transfer the mixture to the wells of a Silicon- $A^{TM}$  Plate<sup>4</sup> on a Collection Plate. Centrifuge at 2,500 5,000 x g for 5 minutes.
- 4. Add 200 μl of **DNA Pre-Wash Buffer** to each well and centrifuge at 2,500 5,000 *x g* for 5 minutes. Discard the flow through.
- Add 300 μl of g-DNA Wash Buffer to each well and centrifuge at 2,500 -5,000 x g for 5 minutes.
- 6. Transfer the Silicon-A<sup>TM</sup> Plate onto an Elution Plate. Add  $\geq$  30  $\mu$ l DNA Elution Buffer or water<sup>3,4</sup> to each well. Incubate 2-5 minutes at room temperature and then centrifuge at 2,500 5,000 x g for 5 minutes to elute the DNA. The eluted DNA can be used immediately for molecular based applications or stored  $\leq$  -20°C for future use.

<sup>&</sup>lt;sup>1</sup>For Proteinase K digested or homogenized material, centrifuge the mixture at 10,000 *x g* for 5 minutes after incubating and transfer up to 600 μl of supernatant to the wells of a Silicon-A<sup>™</sup> Plate on a Collection Plate.

<sup>&</sup>lt;sup>2</sup>The capacity of each well of the Silicon-A™ Plate is ~600 µl.

<sup>&</sup>lt;sup>3</sup>Elution of DNA from the column is dependent on pH and temperature. If water is used, ensure the pH is >6.0. Also, the total yield may be improved by eluting the DNA with Elution Buffer or water pre-equilibrated to 60-70°C.

<sup>&</sup>lt;sup>4</sup>The **DNA Elution Buffer** contains 10 mM Tris-HCl, pH 8.5, 0.1 mM EDTA. If required, pure water can also be used to elute the DNA.

# Proteinase K Digestion with DNA/RNA Shield™

The following protocol requires the additional purchase of **DNA/RNA Shield** (R1100-50 or R1100-250), **Proteinase K w/ Storage Buffer Set** (D3001-2-5 or D3001-2-20) and **PK Digestion Buffer** (R1200-1-5 or R1200-1-20).

#### **Reagent Preparation**

✓ Add 260 µl or 1,040 µl Proteinase K Storage Buffer to reconstitute the lyophilized Proteinase K, 5 mg (D3001-2-5) or 20 mg (D3001-2-20), respectively (final concentration of 20 mg/ml). Vortex to dissolve. Store at -20°C.

#### Protocol

- Add 300 µl of DNA/RNA Shield™ to ≤ 5 mg of solid tissue sample¹. Tissue samples can be mechanically homogenized for optimal extraction efficiency.
- 2. Add 30 µl of PK Digestion Buffer and 15 µl Proteinase K to the sample.
- 3. Mix and then incubate at 55°C until tissue dissolves or up to 5 hours<sup>2</sup>.
- 4. Add 4 volumes of **Genomic Lysis Buffer** to each volume of Proteinase K digestion (4:1). (e.g., add 1,200 μl of Genomic Lysis Buffer to 300 μl of digestion). Mix briefly by vortexing, then let stand at room temperature for 5-10 minutes. Centrifuge the mixture at ≥ 10,000 x g for 5 minutes.
- 5. Transfer the mixture to the wells of a Silicon-A™ Plate³ on a Collection Plate. Centrifuge at 2,500 5,000 x g for 5 minutes. Discard the flow through and repeat this step with the remaining supernatant.
- 6. Add 200  $\mu$ l of **DNA Pre-Wash Buffer** to each well and centrifuge at 2,500 5,000 x g for 5 minutes.
- Add 300 μl of g-DNA Wash Buffer to each well and centrifuge at 2,500 -5,000 x g for 5 minutes.
- 8. Transfer the Silicon-A<sup>TM</sup> Plate onto an Elution Plate. Add  $\geq$  30  $\mu$ l DNA Elution Buffer or water<sup>3,4</sup> to each well. Incubate 2-5 minutes at room temperature and then centrifuge at 2,500 5,000 x g for 5 minutes to elute the DNA. The eluted DNA can be used immediately for molecular based applications or stored  $\leq$  -20°C for future use.

<sup>&</sup>lt;sup>1</sup>Solid Tissue Samples should be completely submerged in **DNA/RNA Shield™**, add as needed.

<sup>&</sup>lt;sup>2</sup>Optimal incubation times may vary with tissue type and homogenization method.

<sup>&</sup>lt;sup>3</sup> The capacity of each well of the Silicon-A™ Plate is ~600 µl.

<sup>&</sup>lt;sup>3</sup> Elution of DNA from the column is dependent on pH and temperature. If water is used, ensure the pH is >6.0. Also, the total yield may be improved by eluting the DNA with Elution Buffer or water pre-equilibrated to 60-70°C.

<sup>&</sup>lt;sup>4</sup>The DNA Elution Buffer contains 10 mM Tris-HCl, pH 8.5, 0.1 mM EDTA. If required, pure water can also be used to elute the DNA.

# **Troubleshooting**

Problem	Possible Causes and Suggested Solutions
DNA degradation	Check for DNase contamination. All reagents supplied with the <i>Quick</i> -DNA™ 96 Kit are DNase-free. However, DNase contamination could result during the processing of some samples. Check pipets, pipet tips, microcentrifuge tubes, etc., and exercise the appropriate precautions during the DNA purification procedure.
DNA is not performing well in subsequent experiments.	Ensure the correct volume of <b>Genomic Lysis Buffer</b> has been added to the sample. Also, make sure all centrifugation steps are completed for the indicated times and speeds (rcfs). Failure to do so may result in incomplete washing, which may cause salts to be eluted with the DNA affecting quantitation and subsequent experiments including enzymatic processes like PCR.
RNA contamination	The buffers in this kit are designed to efficiently hydrolyze and remove RNA during the DNA purification procedure.

# **Ordering Information**

Product Description	Catalog No.	Size
Quick-DNA™ 96 Kit	D3010 D3011 D3012	2 x 96 preps. 4 x 96 preps. 10 x 96 preps.

Individual Kit Components	Catalog No.	Amount
Genomic Lysis Buffer	D3004-1-50 D3004-1-100	50 ml 100 ml
DNA Pre-Wash Buffer	D3004-5-15 D3004-5-30 D3004-5-50	15 ml 30 ml 50 ml
g-DNA Wash Buffer	D3004-2-50 D3004-2-100	50 ml 100 ml
DNA Elution Buffer	D3004-4-10	10 ml
Silicon-A™ Plate	C2001	2 plates
Collection Tubes	C2002	2 plates
Elution Plate	C2003	2 plates

# **Complete Your DNA Methylation Workflow**

#### ✓ Rapid Method for Complete Bisulfite Conversion of DNA

EZ DNA Methylation Kits	Size	Catalog No.
EZ DNA Methylation-Lightning Kit	50 Rxns. 200 Rxns.	D5030 D5031
EZ-96 DNA Methylation-Lightning Kit	2x96 Rxns. (Deep-Well) 2x96 Rxns. (Shallow-Well)	D5032 D5033
EZ DNA Methylation-Lightning Automation Kit	96 Rxns.	D5049
EZ-96 DNA Methylation Lightning MagPrep	4 X 96 Rxns. 8 X 96 Rxns.	D5046 D5047

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Library Prep Kits	Size	Catalog No.
Zymo-Seq WGBS Library Kit	24 Preps.	D5465
Pico Methyl-Seq Library Prep Kit	10 Preps. 25 Preps.	D5455 D5456
Zymo-Seq RRBS Library Kit	24 Preps. 48 Preps.	D5460 D5461

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ZymoTaq Polymerase	Size	Catalog No.
ZymoTaq Premix	50 Rxns. 200 Rxns.	E2003 E2004
ZymoTaq DNA Polymerase	50 Rxns. 200 Rxns.	E2001 E2002
ZymoTaq qPCR Premix	50 Rxns. 200 Rxns.	E2054 E2055

#### ✓ Industry Leading Tools for Assessing Your DNA Methylation Workflow

DNA Methylation Standards	Size	Catalog No.
Human Methylated & Non-methylated DNA Set	5 μg/20 μl	D5014
Universal Methylated DNA Standard	Human Mouse	D5011 D5012
Bisulfite-Converted Universal Methylated Human DNA Standard	1 μg/50 μl	D5015
Human Methylated & Non-Methyated (WGA) DNA Set	5 μg/20 μl	D5013

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