YOUR LEADING SAMPLE PREPATION SOLUTION

FastPrep[®]-24 System & Kits The Most Advanced, Rapid and Thorough Extraction of DNA, RNA and Proteins



FastPrep[®]-24 INSTRUMENT - BEST!

Ultra-Rapid, Thorough Sample Homogenization

Lyse Any Tough or Frozen Sample In 40 Seconds



The FastPrep[®]-24 from MP Biomedicals features a programmable memory of up to five of your most-used settings. The most recent evolution of the famous highthroughput sample preparation system from Bio101[®] Systems, with a total of more than 6,000 installations worldwide, FastPrep provides a unique means of homogenizing by which any type of sample, no matter how tough, can be quickly and consistently lysed within 40 seconds.

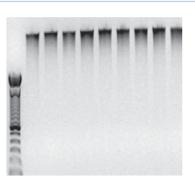
When compared to traditional homogenization methods such as vortexing, syringe shearing, grinding with a mortar and pestle or hammering samples that have been frozen in liquid nitrogen, the FastPrep-24 homogenizer will save hours of work during the sample preparation stage and will provide higher yields of intact DNA, RNA and proteins.

FastPrep[®]-24 uses a unique, optimized motion to disrupt cells through the multidirectional, simultaneous beating of specialized Lysing Matrix beads on the sample material. FastPrep will homogenize up to 24 samples in 2mL tubes, or with optional adapters, lyse 48 samples in 2mL tubes, 12 samples in 15mL tubes or 2 samples in 50mL tubes making FastPrep the most versatile homogenizer available!

Developed for difficult and resistant samples, FastPrep[®]-24 thoroughly and quickly lyses all tissues and cells providing easy and reproducible isolation of stable RNA, active proteins and full-length genomic DNA.

A completely self-contained system, FastPrep[®]-24 eliminates the risk of cross-contamination and timeconsuming clean-up associated with manual lysis methods. Simply add sample and buffers to the Lysing Matrix tube containing specialized beads specific to your application. The ergonomic design of FastPrep ensures ease in loading sample tubes, which remain securely sealed during processing. Homogenization speed and duration times are digitally controlled for consistent results. Upon setting the speed and time with the touch of the a button, simply select this as a memory setting or push "run", and in less than a minute your samples are completely lysed! The new FastPrep®-24 features up to 5 memory presets for your mostused settings.

The unique, vertical angular motion of FastPrep[®]-24 causes the lysing matrix particles to impact the sample from all directions simultaneously, releasing nucleic acids and proteins into the protective buffer faster than with any other system.



Genomic DNA from human ovarian tissue lysed with the FastPrep®-24 for 20 sec. *Courtesy of Dr. David Smith, Oncotech Inc.*

After centrifugation, the supernatant is collected for further purification processing.

A wide variety of specialized Lysing Matrix tubes containing beads of different materials, sizes and shapes have been tailored to guarantee thorough homogenization of samples from such diverse sources as bacteria, yeast, fungi, botanical samples (including seeds), insects, mammalian tissues, (bone, skin, brain, tumor) and cultured cells.

High performance FastPrep Purification Kits, when used in conjunction with the FastPrep[®]-24 Instrument, provide simple, ready-to-use methods for the release and subsequent purification of intact DNA, RNA, and proteins from virtually any source.

Visit www.mpbio.com/sampleprep for additional information or to request a demonstration of FastPrep[®]-24 in your lab.



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FastPrep[®]-24 INSTRUMENT - BEST!

Most Advanced, Rapid and Thorough Extraction of DNA, RNA and Proteins



- Homogenize resistant samples with ease
- Only system with interchangeable adapters allows you to process 24 x 2 mL; 24 x 4.5 mL; 48 x 2 mL; 6 x 15 mL; 12 x 15 mL; or 2 x 50 mL samples
- High reproducibility with precise setting of lysis time and speed. Now available with 5 pre-set memory settings
- Eliminate cross contamination with single-use matrix tubes
- Complete sample preparation for extraction and purification of DNA, RNA and Proteins with FastPrep Kits



EASY-TO-USE PROGRAMMABLE INSTRUMENT!

Save up to 5 pre-set parameters that you use most often, with specific combinations of speed and time.

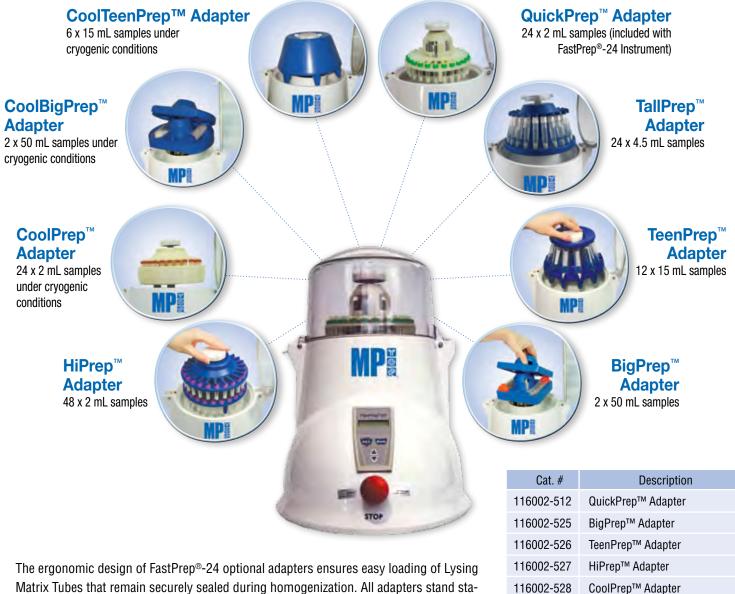
Cat. #	Description	Price
116004-500	FastPrep [®] -24 Instrument	\$9,990.00

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FastPrep[®]-24 INSTRUMENT - BEST!

FastPrep[®]-24 is the ONLY Homogenizer with Interchangeable Adapters!



Matrix Tubes that remain securely sealed during homogenization. All adapters stand stable on the benchtop and are commonly used as tube racks for sample storage at -20°C or -80°C. Frozen Lysing Matrix Tubes loaded in the adapters are ready to be immediately processed in the FastPrep®-24 with minimal hands-on manipulation. Any of our CoolPrep Adapters keep frozen samples chilled to -80°C.

2p 116002-531 116002-540

116002-530

CoolTeenPrep[™] Adapter

CoolBigPrep[™] Adapter

TallPrep[™] Adapter



4

New Cryogenic ADAPTERS - Versatile!

Ideally suited for extractions of any temperature-unstable or sensitive biological compounds such as RNA, siRNA, metabolites, intermediates, and enzymes from even the hardest samples to lyse. Allowing simultaneous cryogenic lysis, the cool adapters ensure efficient cooling of samples through passive temperature control technology with dry ice. Due to high heat transfer capacity and precise settings of lysis parameters, samples can be repeatedly homogenized without any increase in temperature.



CoolPrep[™] 24 x 2 mL Adapter

Cryogenic adapter that holds 24 x 2 mL Lysing Matrix tubes with a tray to hold dry ice, ensuring efficient cooling of samples.

Cat. #	Description	Price
116002-528	CoolPrep [™] Adapter	\$2,750.00



CoolTeenPrep[™] 6 x 15 mL Adapter

Cryogenic adapter that holds 6 x 15 mL Lysing Matrix tubes with a tray to hold dry ice, ensuring efficient cooling of samples.

Cat. #	Description	Price
116002-530	CoolTeenPrep™ Adapter	\$2,750.00



CoolBigPrep[™] 2 x 50 mL Adapter

Cryogenic adapter that holds 2 x 50 mL Lysing Matrix tubes with a tray to hold dry ice, ensuring efficient cooling of samples.

Cat. #	Description	Price
116002-531	CoolBigPrep [™] Adapter	\$2,750.00

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Large-Scale and High Throughput ADAPTERS - Versatile!

Our **large sample volume adapters** are ideally suited for DNA and RNA isolation, enzyme isolation and protein production, natural products isolation, food preparation for quality analysis, biopharma manufacturing, and forensic applications. A wide range of disposable 15 mL and 50 mL Lysing Matrix tubes ensure thorough homogenization of any sample type in seconds.



TeenPrep[™] 12 x 15 mL Adapter

Medium-size adapter that holds 12 x 15 mL Lysing Matrix tubes (compatible with SafTest[™] Food Inspection System).

Cat. #	Description	Price
116002-526	TeenPrep™ Adapter	\$1,250.00



BigPrep[™] 2 x 50 mL Adapter

Large-scale adapter that holds 2 x 50 mL Lysing Matrix tubes (compatible with SafTest™ Food Inspection System).

Cat. #	Description	Price
116002-525	BigPrep™ Adapter	\$1,400.00

Our **high-throughput adapters** are ideally suited for high-throughput applications, up to 24 and 48 samples can be homogenized simultaneously. Additionally, frozen samples in Lysing Matrix tubes loaded in the adapters and stored at -20°C or -80°C are ready to be immediately processed with minimal hands-on manipulation, preventing degradation of cellular components by endogenous enzymes.



HiPrep[™] 48 x 2 mL Adapter

Double decker adapter that holds 48 x 2 mL Lysing Matrix tubes. The ergonomic design of the adapter ensures easy loading of Lysing Matrix tubes that remain securely sealed during the homogenization. It stands stable on the bench top and is commonly used as a tube rack.

Cat. #	Description	Price
116002-527	HiPrep [™] Adapter	\$2,125.00



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TallPrep[™] 24 x 4.5 mL Adapter

Standard adapter that holds 24 x 4.5 mL Lysing Matrix tubes. The ergonomic design of the adapter ensures easy loading of Lysing Matrix tubes that remain securely sealed during the homogenization. It stands stable on the bench top and is commonly used as a tube rack.

Cat. #	Description	Price
116002-540	TallPrep™ Adapter	\$1,200.00

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Lysing Matrix TUBES - Efficient!

MP Biomedicals guarantees the BEST performance from your FastPrep[®]-24 Instrument when used in combination with FastPrep[®] Lysing Matrix Tubes

Matrices are critical components of the FastPrep[®] Sample Preparation system.

Matrices are available separately for use with your own unique buffers, and are also available as components of the complete purification kits on the following pages.



Easy Dispenser Box for Matrix Kits of 100 and 500 Tubes

Cat. #	Description	Pack size	Price
116910-050	Lysing Matrix A	50 x 2 mL Tubes	\$140.70
116910-100		100 x 2 mL Tubes	\$223.10
116910-500		500 x 2 mL Tubes	\$1,044.75
116911-050		50 x 2 mL Tubes	\$140.70
116911-100	Lysing Matrix B	100 x 2 mL Tubes	\$223.10
116911-500		500 x 2 mL Tubes	\$1,044.75
116912-050	Lysing Matrix C	50 x 2 mL Tubes	\$143.40
116912-100		100 x 2 mL Tubes	\$227.35
116912-500		500 x 2 mL Tubes	\$1,064.65
116913-050		50 x 2 mL Tubes	\$140.70
116913-100	Lysing Matrix D	100 x 2 mL Tubes	\$223.10
116913-500		500 x 2 mL Tubes	\$1044.75
116914-050		50 x 2 mL Tubes	\$140.70
116914-100	Lysing Matrix E	100 x 2 mL Tubes	\$223.10
116914-500		500 x 2 mL Tubes	\$1,044.75
116750-200	BioPulverizer™ System I (10 tubes each of 6910-6914)	50 x 2 mL Tubes	\$143.40



Lysing Matrix A

Each impact-resistant 2 mL tube contains garnet matrix and one 1/4 inch ceramic sphere. Extra 1/4 inch ceramic spheres are packaged separately. Lysing Matrix A tubes have orange caps and are found in the FastDNA[®] and FastDNA[®] SPIN Kits. Lysing Matrix A is used for all sample types except soil for the subsequent isolation of genomic DNA.

Lysing Matrix B

Each impact-resistant 2 mL tube contains 0.1 mm silica spheres. Lysing Matrix B tubes have blue caps and are found in the FastRNA[®] Pro Blue Kit and FastProtein[™] Blue Matrix. Lysing Matrix B is used for lysis of gram positive and gram negative bacteria.

Lysing Matrix C

Each impact-resistant 2 mL tube contains 1 mm silica spheres. Lysing Matrix C tubes have red caps and are found in the FastRNA Pro Red Kit and FastProtein Red Matrix. Lysing Matrix C is used for lysis of yeast and fungi.

Lysing Matrix D

Each impact-resistant 2 mL tube contains 1.4 mm ceramic spheres. Lysing Matrix D tubes have green caps and are found in the FastRNA® Pro Green Kit for isolation of total RNA from plants and animals.

Lysing Matrix E

Each impact-resistant 2 mL tube contains 1.4 ceramic spheres, 0.1 mm silica spheres, and one 4 mm glass bead. Lysing Matrix E tubes have purple caps and found in the FastDNA® SPIN Kit for Soil and the FastRNA®Pro Soil Kits.

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TUBES - Efficient!

50 mL Lysing Matrix Tubes



Cat. #	Description	Pack size	Price
116950-010	BigPrep Lysing Matrix A	10 x 50 mL Tubes	\$273.30
116950-050	BigPrep Lysing Matrix A	50 x 50 mL Tubes	\$1,366.50
116951-010	BigPrep Lysing Matrix B	10 x 50 mL Tubes	\$159.00
116951-050	BigPrep Lysing Matrix B	50 x 50 mL Tubes	\$795.00
116953-010	BigPrep Lysing Matrix D	10 x 50 mL Tubes	\$260.00
116953-050	BigPrep Lysing Matrix D	50 x 50 mL Tubes	\$1,050.00
116960-010	BigClean - Lysing Matrix Tubes with stainless steel beads	10 x 50 mL Tubes	\$260.00
116960-050	BigClean - Lysing Matrix Tubes with stainless steel beads	50 x 50 mL Tubes	\$1,050.00
116954-010	BigPrep Lysing Matrix E	10 x 50 mL Tubes	\$159.00
116954-050	BigPrep Lysing Matrix E	50 x 50 mL Tubes	\$795.00

15 mL Lysing Matrix Tubes



Cat. #	Description	Pack size	Price
116930-050	TeenPrep Lysing Matrix A	50 x 15 mL Tubes	\$562.80
116931-050	TeenPrep Lysing Matrix B	50 x 15 mL Tubes	\$562.80
116932-050	TeenPrep Lysing Matrix C	50 x 15 mL Tubes	\$562.80
116933-050	TeenPrep Lysing Matrix D	50 x 15 mL Tubes	\$562.80
116934-050	TeenPrep Lysing Matrix E	50 x 15 mL Tubes	\$562.80

FastPrep[®]-24 Specifications



Time:

1-60 seconds programmable in 1 sec. increments

Speed:

4.0 - 6.5 m/s in programmable in 0.5 m/s increments

Acceleration: <2 sec. to max speed

Deceleration:

<2 sec. to stop

Standard Environmental Temperature Operating Range: 4-40 °C

Dimensions: Height - 270mm; Base (Ellipse) - 425 - 330mm

Weight: 17.5 kg/45 lbs

7.5 Kg/45 IDS

Power Requirements: 90-250 V AC, 50/60 Hz, 1,200 W

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Isolation and Purification SYSTEMS - Complete!

Ready-to-Use Protocols for DNA, RNA and Protein Isolation from Any Sample! A Wide Range of FastPrep® Kits

- Rapid and reproducible sample lysis and purification process
- No cross contamination with closed lysing matrix tubes
- Increased yields of high quality DNA, RNA and Proteins
- Integrity and size of DNA, RNA and Proteins are retained
- Nucleic acids and Proteins are ready-to-use in downstream applications

FastProtein[™] Blue Matrix

Release of proteins from gram positive and gram

· Protein extracts are ready for immediate electro-

Cat N° 6550-400 (50 preps) - Cat N° 6550-500 (100 preps)

· Ideal for optimizing induction conditions

RNA

negative bacteria in 40 seconds

phoresis or purification

NEW

FastDNA[™] Spin Kit for Soil

DNA

Cat N° 6560-200 (50 preps)

- Variety of soil and environmental sample types
- No hazardous organic reagents required
- SPIN filters streamline silica handling

FastDNA[™] Spin Kit for Feces

Cat N° 6570-200 (50 preps)

- Removes organic contaminates, such as Humic acid, for downstream applications
- Achieves Optimal 260/280 ratios (1.8-2.0)
- Typically isolates 10–20 µg of gDNA from 500 mg of stool

FastRNA[™] Pro Green Kit

Cat N° 6045-050 (50 preps)

- For use with all plant and animal samples
- Lyse 50-100 mg tissue per 2 mL tube

FastRNA[™] Pro Soil-Direct Kit FastRNA[™] Pro Soil-Indirect Kit

Cat N° 6070-050 - Cat N° 6075-050 respectively (50 preps)

- Variety of soil and environmental sample types
- RNA protected during and after processing
- Humic acids reduced to allow uninhibited RT-PCR
- Includes additional reagents for even further purification if necessary
- SPIN filters streamline silica handling

PROTEIN

FastPrep[®]

Kits

FastRNA[™] Pro Red Kit

Cat N° 6035-050 (50 preps)

- For use with yeast cells and fungal tissue
- Lyse up to 10¹⁰ cells per 2 mL tube

FastRNA[™] Pro Blue Kit

Cat N° 6025-050 (50 preps)

- For use with gram positive and gram negative bacteria
- Lyse up to 10¹⁰ cells per 2 mL tube

FastDNA[™] Kit FastDNA[™] Spin Kit

Cat N° 6540-400 - Cat N° 6540-600 respectively (100 preps)

- Plant, animal, yeast, fungal and microbial samples
- No hazardous organic reagents required
- SPIN filters streamline silica handling (FastDNA Spin Kit)

FastProtein[™] Red Matrix

Cat N° 6550-600 (50 preps) - Cat N° 6550-700 (100 preps)

- Release of proteins from yeast cells and fungi in 40 seconds
- Protein extracts are ready for immediate electrophoresis or purification
- Ideal for optimizing induction conditions

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Isolation and Purification SYSTEMS - Proven!

FastDNA[™] SPIN Kit

A Rapid Method of Isolating Pure Genomic DNA from a Wide Variety of Sources!

- Rapid and reproducible sample lysis with the FastPrep[®]-24 or FastPrep[®] FP120
- Isolate PCR-ready DNA from a variety of sample types
- No hazardous organic reagents are required

The FastDNA[™] SPIN Kit quickly and efficiently isolates genomic DNA from almost any sample (plant and animal tissues, cultured cells, bacteria, yeast, fungi, insects, etc). Up to 200 mg of tissue or cells are processed by the FastPrep®-24 or FastPrep® FP120 with the Lysing Matrix A tubes. The kit includes 3 different chaotropic buffers for the homogenization of a wide variety of sample types and the released DNA is purified by a silica-based spin filter method. Purified DNA is ready for enzyme digestion, electrophoresis, PCR and any other desired application.

References

1. Hill J.E. et al (2005). Appl.Environ.Microbiol. Vol 71 : 867-875

2. Moon H. et al (2004). *J.Exp.Bot.* Vol 55 : 1519-1528 3. Diopici H.M. et al (2004). *Appl Envir Microbiol.* Vol 70 : 3988-3995

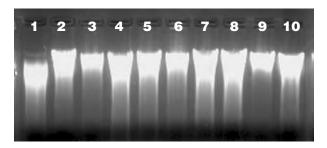
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Cat #	Designation	Pack Size	Price
116540-600	FastDNA™ SPIN Kit	100 Preps	\$344.20

FastDNA[™] SPIN Kit For Soil Isolate Pure DNA from Cells Present in Soil or Other Environmental Samples!

- Rapid and reproducible sample lysis with the FastPrep[®]24 or FastPrep[®] FP120 Instrument
- Easily isolate DNA from a variety of organisms in many different types of soil
- No hazardous organic reagents are required

The FastDNA[™] SPIN Kit for Soil is designed to efficiently isolate bacterial, fungi, plant and animal genomic DNA from soil and environmental samples. Up to 500 mg soil are processed by the FastPrep[®]-24 or FastPrep[®] FP120 with the Lysing Matrix E tubes designed to efficiently lyse all microorganisms including difficult sources such as eubacterial spores and endospores, gram positive bacteria and yeast. The released DNA is purified by a silica-based spin filter method and is suitable for PCR analysis and other downstream applications.



DNA from various soil samples extracted with the FastDNA® SPIN Kit for Soil.

20% of the DNA isolated from 500 mg soil was loaded on a 1.2% agarose gel (0.5X TAE).

Soil was taken from:	
Lane 1: tomato pot;	Lane 2 : sludge
Lane 3 : sandy soil;	Lane 4 : under pine tree
Lane 5 : under palm tree;	Lane 6 : green garden
Lane 7 : Nile Lilly pot;	Lane 8 : lawn grass
Lane 9 : citrus tree;	Lane 10 : avocado tree. DNA ranges from 4-20 kb.

References

- 1. Selesi D. et al (2005). Appl. Envir. Microbiol. Vol 71 : 175-184
- 2. Alexandrino M. et al (2004). Water Research. Vol 38 : 1340-1346
- 3. Mumy K.L. et al (2004). J. of Microbiological Methods. Vol 57 : 259–268

Cat #	Designation	Pack Size	Price
116560-200	FastDNA [™] SPIN Kit for Soil	50 Preps	\$240.45



Isolation and Purification SYSTEMS - Proven!

FastDNA SPIN[™] Kit for Feces Isolate High Quality DNA from Fecal Material

- Rapid and reproducible sample lysis in under 40 seconds with the FastPrep®-24
- Removes organic contaminates, such as Humic acid, for downstream applications
- Achieves Optimal 260/280 ratios (1.8-2.0)
- Typically isolates 10 μg 20 μg of gDNA from 500 mg of stool

The FastDNA[™] Spin Kit for Feces is the newest addition to the evolving FastDNA[™] kit family. Prompted by you, our customer, MP Biomedicals has developed a FastDNA[™] Spin Kit designed exclusively for the isolation of genomic DNA from fecal material. With the FastDNA[™] Spin Kit for Feces you will have everything you need to quickly and efficiently lyse any fecal sample isolating high quality DNA for immediate use in downstream applications. Used in conjunction with our FastPrep[®]-24 homogenization system you will be able to completely lyse fecal samples in seconds with no pre-grinding or preparation.

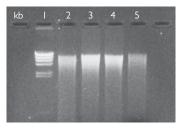
Cat #	Designation	Pack Size	Price
116570-200	FastDNA [™] SPIN Kit for Feces	50 Preps	\$240.45

FastRNA[™] Pro Kits Isolate High Quality Total RNA with a Single-Reagent Extraction Method!

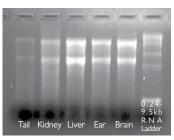
- Rapid and reproducible sample lysis in under 40 seconds with the FastPrep[®]24 or FastPrep[®] FP120
- Safe and consistent RNA isolation with the single-reagent RNAPro™ solution
- Lysis and purification of total RNA

The FastRNA[™] Pro Kits are designed to quickly and efficiently isolate total RNA from virtually any sample. During the homogenization step, intact total RNA is released in the proprietary RNAPro solution where it is immediately stabilized. The RNAPro solution inactivates cellular RNases during cell lysis to prevent RNA degradation. RNA is then extracted with chloroform and precipitated with ethanol. DEPC-treated water is provided for resuspension of total RNA. High quality RNA prepared with FastRNA[™] Pro Kits is ready for all downstream applications including RT-PCR, gene expression and microarray analysis.

Cat #	Designation	Pack Size	Price
116025-050	FastRNA™ Pro Blue Kit (bacteria)	50 Preps	\$145.55
116035-050	FastRNA™ Pro Red Kit (yeast and fungi)	50 Preps	\$148.30
116045-050	FastRNA™ Pro Green Kit (plants and animals)	50 Preps	\$145.55



DNA from fecal samples with the FastDNA™ Spin Kit for Feces. DNA was loaded on a 1.2% agarose gel (0.5X TAE). Lamda HindIII Marker Bovine stool 200ng DNA Equine stool 200ng DNA Feline stool 200ng DNA Avian stool 200ng DNA



Rat total RNA extracted with the FastRNA® Pro Green Kit. Approximatively 2% of the total RNA isolated from 100 mg frozen tissue was loaded on to a 1.2% denaturing agarose gel (1X MOPS).

References

- 1. Vido K. et al (2005). *J. Bacteriol.* Vol 187 : 601-610 2. Tsai H.F. et al (2004). *Antimicrob.*
- Agents Chemoth. Vol 48 : 2483 2489
- 3. Tupin E. et al (2004). J. Exp. Med. Vol 199 : 417 -422

FastRNA[™] Pro Soil Kits Isolate Total RNA From Soil that is Immediately Ready for RT-PCR and other Downstream Applications!

- Rapid and reproducible sample lysis in under 40 seconds with the FastPrep[®]-24 or FastPrep[®] FP120
- Easily lyse difficult gram positive cells, plant material, and organic debris directly from soil
- Lysis and purification solutions protect RNA during processing
- Humic acids levels reduced to allow uninhibited RT-PCR
- Lysis and purification of total RNA

The FastRNA[™] Pro Soil-Direct and Indirect kits are designed to efficiently isolate total RNA from organic material found in soil samples or soil supernatants. The direct method consists of extracting nucleic acid from microorganisms and other biological specimens directly from soil. The indirect method utilizes an initial separation of microorganisms and other biological specimens from the soil followed by lysis of the organisms and RNA purification. This method also permits soil incubation with growth media in order to amplify under-represented living organisms prior to RNA isolation if specific comparisons of microbial diversity are not desired. FastRNA[™] Pro Soil kits purify RNA in a process that removes humic substances and other inhibitors, and efficiently inactivates cellular RNases during homogenization to prevent RNA degradation. Purified RNA is thus suitable for RT-PCR analysis and other downstream applications.

Cat #	Designation	Pack Size	Price
116070-050	FastRNA™ Pro Soil-Direct Kit	50 Preps	\$360.60
116075-050	FastRNA™ Pro Soil-Indirect Kit	50 Preps	\$360.60

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Isolation and Purification SYSTEMS - Proven!

Easy Lysis of Microorganisms to Release Recombinant Proteins

FastProtein[™] Matrix

- Save time by reducing sample lysis time to seconds
- Quickly and consistently lyse samples from different time points or induction conditions
- Protein extract is ready for immediate electrophoresis or purification

Prepare dozens of protein samples in minutes!

The FastProtein[™] products employ a powerful, patented technology for the rapid lysis of yeast and bacteria. Used in conjunction with the FastPrep[®]-24 or FastPrep[®] FP120 Instrument, these products offer the fastest way to release expressed proteins from the host organism. FastProtein[™] Kits are perfect for analyzing protein expression conditions using gel analysis. Samples are enclosed during the quick lysis step, thus preventing cross-contamination or sample loss.

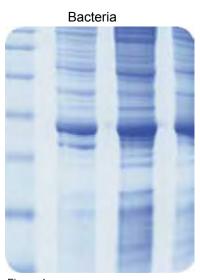


Figure 1 12% SDS PAGE of lysate of BL21 cells expressing the GST protein resulting from homogenization with the FastProtein[™] Blue Matrix

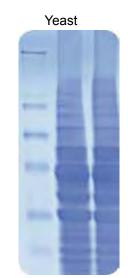


Figure 2 12% SDS PAGE of lysate of yeast cells resulting from homogenization with FastProtein™ Red .

FastProtein[™] Blue For Lysis Of Gram Positive And Gram Negative Bacteria



The FastProtein[™] Blue matrix is optimal for lysing gram positive and gram negative bacteria. These fine glass beads are designed for use with gram positive bacteria or any difficult microorganism. Cells, resuspended in either 1X PBS or your own expression buffer, are added to the Lysing Matrix and processed in the FastPrep[®]-24 or FastPrep[®] FP120 for 20-40 seconds.

Cat. #	Description	Size	Price
116550-400	FastProtein™ Blue Matrix	50 x 2 mL	\$143.40
116550-500	FastProtein™ Blue Matrix	100 x 2 mL	\$223.10
116550-600	FastProtein™ Red Matrix	50 x 2 mL	\$144.45
116550-700	FastProtein™ Red Matrix	100 x 2 mL	\$228.65

FastProtein[™] Red For Lysis Of Yeast Cells

The FastProtein[™] Red matrix is used to lyse yeast cells. Cells, resuspended in either Yeast Breaking Buffer (YBB-supplied with the kit) or your own expression buffer, are added to the small glass beads of this Lysing Matrix and processed in the FastPrep[®]-24 or FastPrep[®] FP120 Instrument for 20-40 seconds.

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FastPrep® SYSTEM SETTINGS

Typical FastPrep® System Settings for Optimal Sample Lysis

Sample Name	Sample Type	Quantity	Lysing Matrix	FastPrep [®] speed	FastPrep® time	
	HUMAN AND ANIMAL					
Human	Lung	50 mg	Lysing Matrix D	6.0	4 x 30 sec.	
Human	Breast	80 mg	Lysing Matrix D	6.0	2 x 30 sec.	
Human	Kidney	50 mg	Lysing Matrix D	6.0	40 sec.	
Human	Thyroid Tumors	100 mg	Lysing Matrix A	6.0	3 x 30 sec.	
Mouse	Eye	10 mg	Lysing Matrix D	6.0	4 x 30 sec.	
Mouse	Heart	70 mg	Lysing Matrix D	6.0	4 x 30 sec.	
Mouse	Kidney	50 mg	Lysing Matrix D	6.0	40 sec.	
Mouse	Femur	40 mg	Lysing Matrix A	6.0	4 x 30 sec.	
Mouse	Leg Muscle	50 mg	Lysing Matrix D	6.0	40 sec.	
Mouse	Intestine	50 mg	Lysing Matrix D	6.0	40 sec.	
Mouse	Ear	45 mg	Lysing Matrix D	6.0	4 x 30 sec.	
Mouse	Tail	100 mg	Lysing Matrix A	6.0	4 x 30 sec.	
Mouse	Spleen	70 mg	Lysing Matrix D	6.0	40 sec.	
Mouse	Lung	50 mg	Lysing Matrix D	6.0	40 sec.	
Mouse	Liver	50 mg	Lysing Matrix D	6.0	40 sec.	
Mouse	Brain	50 mg	Lysing Matrix D	6.0	40 sec.	
Mouse	Pancreatic cells (bHC9)	10 ⁷ cells	Lysing Matrix D	6.0	40 sec.	

		PLANT			
Alpowa Wheat	Leaf Tissue	75 mg	Lysing Matrix D	6.0	40 sec.
Alpowa Wheat	Seed	100 mg	Lysing Matrix A	6.0	40 sec.
Arabidopsis thaliana	Fresh Leaves	50 mg	Lysing Matrix D	6.0	40 sec.
Arabidopsis thaliana	Fresh Leaves	200 mg	Lysing Matrix D	6.0	2 x 40 sec.
Bartlett Pear	Leaf Tissue	50 mg	Lysing Matrix D	6.0	40 sec.
Classic Oat	Leaf Tissue	75 mg	Lysing Matrix D	6.0	40 sec.
Classic Oat	Seed	100 mg	Lysing Matrix A	6.0	40 sec.
Corn	Leaf Tissue	100 mg	Lysing Matrix D	6.0	40 sec.
Crest Barley	Leaf Tissue	100 mg	Lysing Matrix D	6.0	40 sec.
Crest Barley	Root	300 mg	Lysing Matrix A	6.0	40 sec.
Kaybonnet Rice	Leaf Tissue	100 mg	Lysing Matrix D	6.0	40 sec.
Kaybonnet Rice	Seed	100 mg	Lysing Matrix A	6.0	40 sec.
Klages Barley	Root	300 mg	Lysing Matrix A	6.0	40 sec.
Klages Barley	Leaf Tissue	70 mg	Lysing Matrix D	6.0	40 sec.
Tobacco	Leaf Tissue	75 mg	Lysing Matrix D	6.0	40 sec.

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FastPrep® SYSTEM SETTINGS

Typical FastPrep[®] System Settings for Optimal Sample Lysis

Sample Name	Sample Type	Quantity	Lysing Matrix	FastPrep [®] speed	FastPrep® time
		PLANT			
Lafitte Rice	Leaf Tissue	75 mg	Lysing Matrix D	6.0	40 sec.
Lafitte Rice	Sprout Leaf	100 mg	Lysing Matrix D	6.0	2 x 30 sec.
Soybean	Seed	100 mg	Lysing Matrix A	6.0	40 sec.
Corn	Seed	100 mg	Lysing Matrix A	6.0	40 sec.
Oat FL 502	Leaf Tissue	75 mg	Lysing Matrix D	6.0	40 sec.
Oat FL 502	Seed	100 mg	Lysing Matrix A	6.0	40 sec.
Riser Oat	Leaf Tissue	70 mg	Lysing Matrix D	6.0	40 sec.
Richland Soybean	Leaf Tissue	100 mg	Lysing Matrix D	6.0	40 sec.
Tam Wheat	Leaf Tissue	75 mg	Lysing Matrix D	6.0	40 sec.
Tam Wheat	Root	80 mg	Lysing Matrix A	6.0	40 sec.
Tomato, Early Girl	Leaf Tissue	75 mg	Lysing Matrix D	6.0	4 x 30 sec.
Williams 82 Soybean	Leaf Tissue	70 mg	Lysing Matrix D	6.0	40 sec.
Wrens Rye	Leaf Tissue	100 mg	Lysing Matrix D	6.0	40 sec.
Pine	Needle	100 mg	Lysing Matrix A	6.0	30 sec.
		DAOTED	1.4		
		BACTER			
Listeria monocytogenes	Cells	10 ⁹ cells	Lysing Matrix B	6.0	3 x 30 sec.
Streptococcus pyogenes	Cells	10 ⁹ cells	Lysing Matrix B	6.0	20 sec.
Streptococcus mutans	Cells	10 ⁹ cells	Lysing Matrix B	6.0	30 sec.
Staphylococcus aureus	Cells	10 ⁸ cells	Lysing Matrix B	6.0	2 x 40 sec.
Photorhabdus luminescens	Cells	10 ⁹ cells	Lysing Matrix B	6.0	2 x 30 sec.
Escherischia coli	Cells	10 ⁸ cells	Lysing Matrix B	6.0	30 sec.
Mycobacterium tuberculosis	Cells	10 ⁸ cells	Lysing Matrix B	6.0	2 x 45 sec.
Lactococcus lactis	Cells	10 ⁸ cells	Lysing Matrix B	6.0	3 x 30 sec.
		YEAST AND	FUNGI		
Saccharomyces cerevisiae	Cells	10 ⁸ cells	Lysing Matrix C	6.0	40 sec.
Schizosaccharomyces pombe	Cells	10 ⁸ cells	Lysing Matrix C	5.0	4 x 15 sec.
Candida albicans	Cells	10 ⁸ cells	Lysing Matrix C	6.0	2 x 30 sec.
Cryptococcus neoformans	Cells	10 ⁸ cells	Lysing Matrix C	6.0	4 x 30 sec.
Aspergillus fumigatus	Cells	10 ⁸ cells	Lysing Matrix C	6.0	2 x 30 sec.
Fusarium solani	Cells	10 ⁸ cells	Lysing Matrix C	6.0	2 x 30 sec.
		ENVIRONMENTAL			
Sediments	Soil/rocks	50 mg	Lysing Matrix E	5.5	2 x 30 sec.
Soil	Sandy sample	50 mg	Lysing Matrix E	4.0	4 x 30 sec.
Soil	Litter	50 mg	Lysing Matrix E	5.5	30 sec.
Feces	Turd	300 mg	Lysing Matrix E	6.0	40 sec.

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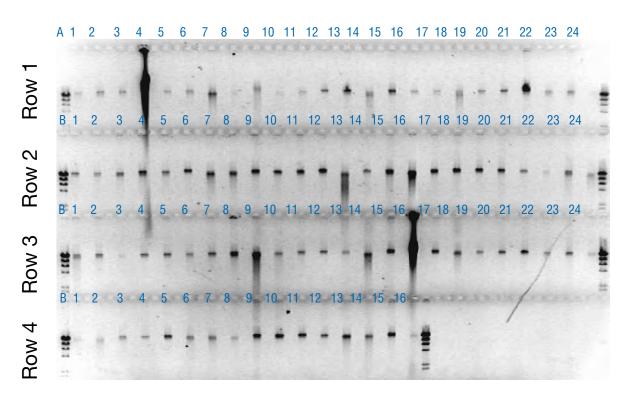
Application OVERVIEW TUMORS

Example of Onco-Pathology Related Applications of FastPrep[®]-24 Sample Preparation of Biopsy Tissues for Genomic Analysis and Drug Resistance Screening



Human-derived biopsy specimens of primary and secondary tumors are usually complex matrices which are very hard to properly homogenize using the classical methods. Their mechanical consistencies vary widely. The FastPrep[®]-24 System, with its unique disruption mechanism and accurate settings, allows for rapid, repeatable and reliable sample lysis and homogenization, and produces highest quality of functional genomic DNA, RNA and proteins for a variety of research, diagnostics and pharmacology applications.

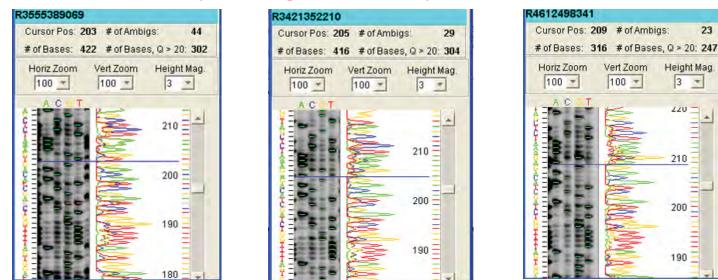
As an example of high-throughput screening, here are 85 genomic DNAs isolated from human ovary cancer tumor biopsies via the FastPrep[®]-24, and automatically purified via the BioMEK3K robot. FastPrep[®] prepared lysate is directly compatible with third party high-throughput automation and automation kits



FastPrep®-24 settings: 6.0 m/s for 40 seconds (one pass), using Lysing Matrix A

Application OVERVIEW TUMORS

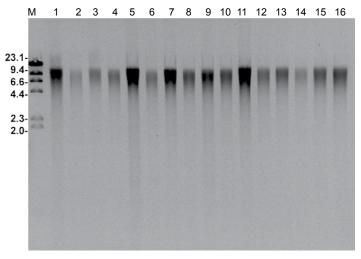
Sequencing of FastPrep® Isolated DNA



The FastPrep[®]-Isolated DNA is Ready for High-Throughput Sequencing

An example of the sequencing analysis of genomic DNA extracted using the FastPrep®-24 system from a single biopsy melanoma tissue. The target wild type sequence is GTGACA with a documented mutation at "T" (usually a tranversion to "A"). Note that the first panel is wild type, second panel is a mixture of wild type and mutant and the third panel is a pure mutant. This shows clear evidence for heterogeneity within a single biopsy specimen. The sequencing gels further demonstrate the high quality of DNA extracted from lysates generated by the FastPrep® System.





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Gel of gDNA isolated from melanoma specimens



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Application OVERVIEW TISSUE

Sample Overview

Animal and human tissues can be the toughest samples to isolate high-quality DNA, RNA, and proteins. Using the FastDNA[™] Spin Kit and FastRNA[™] Pro Green Kit in combination with the FastPrep instrument, full homogenization of any sample including bone and tumors, and more elastic samples like skin, is achieved within a few seconds. This method saves hours of work during sample preparation and provides high yields of DNA, RNA, and proteins. Effective, efficient sample preparation is critical to successful downstream results.

RNA and Protein Extraction From Skin Tissue

The FastPrep and associated matrices have demonstrated successful lysis and dual extraction of RNA and proteins from skin tissue in three runs of 40 seconds each.

Materials

- FastPrep® instrument
- Lysing Matrix D tubes
- Sample: Human skin biopsies from a 3-mm punch, weighing only 19 mg on average

Protocol and Parameters

- 1. Add the skin sample to a Lysing Matrix D tube.
- Add 1 mL of a guanidine thiocyanate lysis buffer (5.1 M guanidine thiocyanate, 50 mM sodium citrate, 50 mM EDTA, 0.5% β-mercaptoethanol).
- 3. Homogenize in the FastPrep[®] instrument for 3 x 40 seconds at a speed setting of 6.0. Place the tubes on ice for 5 minutes between each run.
- 4. Centrifuge at 14,000 x g for 5-10 minutes to pellet debris.
- 5. Proceed with the RNA and protein extraction protocol.

Results

- The average yield of 1.4 μ g RNA obtained with the FastPrep[®] System was 57% higher than yields obtained with the Polytron (Figure 1).
- The average yield of 170 µg protein obtained with the FastPrep[®] System was 53% higher than yields obtained with the Polytron (Figure 1).
- To verify the high-quality nature of the RNA, samples were analysed with the Agilent 2100 Bioanalyzer. samples had ribosomal integrity number numbers of between 8.4 and 8.9, which is consistent with high-quality RNA (Figure 2).
- The quality of extracted proteins was assessed by two-dimensional gel and Western blot analysis. There was distinct spot resolution and sufficient protein isolated from single biopsy to produce five to six two-dimensional gels. For Western blotting, a primary antibody against GADD-45 was used to probe the membrane. GADD-45 antibody detects both the alpha and beta portions of the protein, although it is more sensitive for the alpha portion.

	RNA average quantity per biopsy (lg)	RNA average 260/280 ratio	Protein average quantity per biopsy (lg)
FastPrep Homogenizer	1.4 (±0.4 µg)	2.0 (±0.05)	170 (±50 μg)
Polytron	0.8 (±0.4 µg)	1.8 (±0.11)	90 (±40 µg)

Figure 1. RNA and protein quantitation for each method of tissue disruption, the quantity and quality of RNA (as an OD260/280 ratio) and the quantity of protein is shown. The RNA was quantified using the NanoDrop® spectrophotometer and the protein content was determined using a Bradford-based assay. For RNA, an OD260/280 of 2.0 is optimal. All quantities are ±SD.

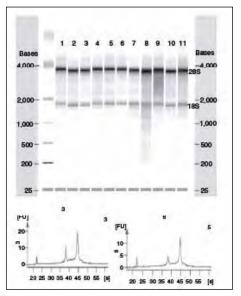


Figure 2. The RNA was run on an Agilent 2100 Bioanalyzer (Agilent, Palo Alto, Calif.) using the RNA 6000 Pico LabChip kit to determine the quality of the samples. The 28S and 18S ribosomal bands show a greater than 2:1 ratio, and the calculated RNA ribosomal integrity numbers of the samples ranged from 8.4 to 8.9, verifying high-quality RNA. Shown above are the gel images for 11 RNA samples and below are two representative electrophoretic graphs showing the RNA peaks.



Application OVERVIEW PLANT

Sample Overview

Because plant samples can be very fibrous and contain high levels of polyphenolic compounds, polysaccharides, and RNases, it can be extremely difficult to extract enough usable DNA or RNA for PCR analysis and other downstream applications. The FastDNA[™] Kit, FastDNA[™] Spin Kit, and FastRNA[™] Pro Green Kit make it fast and simple! Effective, efficient sample preparation is critical to successful downstream results.

RNA Extraction From Cassava Root

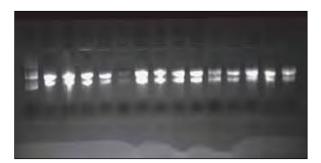
FastPrep and associated matrices have demonstrated successful lysis and RNA extraction from cassava roots in only 60 seconds.

Materials

- FastPrep® instrument
- Lysing Matrix A with an additional 1/4-inch ceramic bead
- Sample: Cassava roots

Protocol and Parameters

- 1. Add the Cassava root sample to a Lysing Matrix A tube containing an additional ¹/₄-inch ceramic bead.
- 2. Add 1 mL of RNApro extraction buffer.
- 3. Homogenize in the FastPrep instrument for 60 seconds at a speed setting of 6.0.
- 4. Centrifuge at 14,000 x g for 5-10 minutes to pellet debris.
- 5. Collect the supernatant and proceed with the RNA extraction protocol.



RNA extraction from Cassava storage roots; samples contain 0.32 $\mu g/\mu$ -110 $\mu g/\mu$ l RNA. FastPrep settings: Speed 6.0 for 60 s; Lysing Matrix A with additional ceramic 1/4-inch bead

Results

 RNA could successfully be extracted from cassava roots with the FastPrep System. Total RNA yields achieved were up to 110 μg of RNA per mL.

Product Overview

The FastDNA[™] Kit and FastDNA[™] Spin Kit are used with the FastPrep[®] instrument to lyse and subsequently isolate DNA from up to 200 µg of almost any plant sample in less than 30 minutes.

The FastRNA™ Pro Green Kit is designed to efficiently isolate total RNA from any type of plant and animal tissue or cultured cells.



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Total RNA extracted from basil leaves with the FastRNATM Pro Green Kit. 50 mg of basil leaves were homogenized with the FastPrep[®] instrument for one, two, or three runs of 40 seconds performed at a speed setting of 6.0. Lane 1: Marker; Lane 2: 1 x 40 sec; Lane 3: 2 x 40 sec; Lane 4: 3 x 40 sec.



Extracted DNA from various plant samples using the FastDNA™ Kit: Approximately 1 µg of isolated DNA was loaded on a 1.2% agarose gel (0.5X TAE). Lane 1: ~0.16 g apple stem; Lane 2: ~0.45 g red bell pepper seeds; Lane 3: ~0.45 g pelagonium root; Lane 4: ~0.45 g mature peace lily leaf; Lane 5: ~0.45 g ice plant leaf; Lane 6: Lambda Hind III marker.



Application OVERVIEW SOIL

Sample Overview

The challenge with extractions from soil is isolating DNA or RNA without contamination by humic acids or other PCR inhibitors. The FastDNA[™] Spin Kit for Soil and FastRNA[™] Pro Soil Kits used in combination with the FastPrep[®] instrument will help overcome any difficulties with complete lysis of all soil organisms including historically difficult sources such as eubacterial spores and endospores, gram positive bacteria, yeast, algae, nematodes and fungi, and isolation of pure DNA and RNA. Effective, efficient sample preparation is critical to successful downstream results.

DNA Extraction From Andisol, a Volcanic Ash Soil

DNA extraction from Andisol, a volcanic ash soil, is known to be very difficult because this soil has a complex matrix, including allophane as a clay mineral. Soil properties such as high clay content contribute to high adsorption of DNA to soil particles. The combination of the FastPrep instrument and the FastDNA[™] SPIN Kit for Soil used together with skim milk have demonstrated successful extraction of PCR-suitable DNA from recalcitrant soil samples like volcanic ash soil.

Materials

- FastPrep[®] instrument
- FastDNA™ Spin Kit for Soil
- Skim milk (carrier minimizing adsorption of nucleic acids to soil)
- Sample: Andisol, volcanic ash soils

Protocol and parameters

- Add the soil sample together with or without 40 mg skim milk per gram of soil to a Lysing Matrix E tube.
- 2. Add 978 μL sodium phosphate buffer to the sample in the Lysing Matrix E tube.
- 3. Add 122 µL MT Buffer.
- 4. Homogenize in the FastPrep instrument for 40 seconds at a speed setting of 6.0.
- 5. Centrifuge at 14,000 x g for 5-10 minutes to pellet debris.
- 6. Follow the FastDNA[™] Spin Kit for Soil protocol for DNA purification from the homogenate.

Results

DNA could successfully be extracted from Andisol soil samples with the FastDNA[™] Spin Kit for Soil and the addition of 40 mg of skim milk per gram of soil sample. PCR products of the expected size were amplified from all extracts with skim milk. Resultant extracts were suitable for PCR and no other purification procedures were needed.

Product Overview

The FastDNA[™] Spin Kit for Soil is designed to efficiently isolate bacteria, fungal, plant, and animal genomic DNA from soil and other environmental samples. The FastRNA[™] Pro Soil Kits are designed to efficiently isolate total RNA from organic material found in soil samples and soil sample supernatants.

Soil no	Origin	Soil taxonomy ^{a)}	Soil texture	pH (H ₂ O)	Organic C content (g kg ⁻¹)	P retention (%)
1	Spinach field, Ibaraki	Dystric-Silic Andisol	light clay	5.46	83.419	83
2	Conserved forest, Ibaraki	Dystric-Silic Andisol	light clay	4.84	149.43	84
3	Apple orchard, Aomori	Silic-Eutrisilic Andisol (Dystric)	sandy clay loam	6.08	122.893	75
4	Vegetable field 1, Fukushima	Dystric-Silic Andisol	light clay	6.20	78.795	71
5	Vegetable field 2, Fukushima	Haplic-Dystric Cambisol	clay loam	6.02	23.239	65
6	Upland crop field, Kumamoto	Dystric-Silic Andisol	heavy clay	5.59	117.283	82
7	Paddy field, Kumamoto	Silic-Eutrisilic Andisol (Dystric)	heavy clay	6.38	119.425	91

a): According to the world reference base (WRB) for soil resources classification.

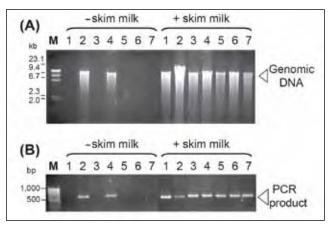


Fig. 2. Agarose gel electrophoresis of DNA and PCR products extracted from sample soils (1-7) FastPrep was used for extracting DNA from soils (A). PCR products from these extracts were amplified with bacterial 16S rONA universal primer set (338f and 907r) (B) when amended with (+) or without (-) 40 mg of skim milk g⁻¹ soil. Numbers show soil samples listed in Table I. M: Molecular marker (A: λ JHind III digest, B: 100 bp ladder).

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FastPrep[®]-24 APPLICATIONS



Questions and Answers

Is it possible to isolate virus particles from animal tissues with the FastPrep[®]-24 System?

The FastPrep[®]-24 System is designed for the isolation of viruses from animal and plant tissues. Researchers using the FastPrep[®]-24 instrument for this application reported that sample homogenization with Lysing Matrix A tubes (garnet sand and one ceramic bead) induce lysis of viruses together with the animal cells. When the garnet sand is removed and the homogenization process is performed with one ceramic bead, the animal cells are lysed after one run of 20 sec. at speed 6.5 m/s but not the viruses, meaning that the protein capsid is intact.

Reference: Klempa B. et al. (2005) *J.Clin.Microbiol*. Vol 43(6); 2756-2763.

Are there specific recommendations to prevent RNA degradation by RNases when isolating RNA with the FastRNA[™] Pro Kits?

The origin of RNA degradation is often action of RNases: both endogenous and exogenous RNases.

1. Inactivation of endogenous RNases

Endogenous RNases are released from cellular compartments immediately after harvesting tissue and cells. It is essential to inactivate these RNases as soon as possible to prevent RNA degradation. To effectively inactivate endogenous RNases, add RNApro Solution (chaotropic-based cell lysis solution containing guanidium isothiocyanate) to each sample as soon as possible following sample harvest and homogenize immediately with the FastPrep®-24 instrument or flash-freeze samples in liquid nitrogen. To prevent RNA degradation, it is important that the tissue be cut in small enough (1 cm) pieces to allow rapid, thorough freezing of the entire tissue.

2. Reduce exposure to exogenous RNases

To isolate intact, high quality RNA, it is essential that exogenous RNases are not introduced into purified RNA preparations. It is essential that any item that could contact the purified RNA is RNase-free. All surfaces, including pipettors, benchtops, glassware and gel equipment, should be decontaminated with a surface decontamination solution such as RNase Erase (Cat # 112440204). RNase-free tips, tubes, and solutions should always be used and gloves should be changed frequently.

What are the settings for yeast lysis with the BigPrep[™] Adapter ?

The FastPrep[®]-24 instrument in combination with the BigPrep[™] Adapter has been successfully used for the lysis of *Pichia pastoris*, a yeast strain that has a big cell wall. After centrifugation of 1 liter culture (5 x 10⁸ cells/ mL), the cell pellet is resuspended in 50 mL Lysis Buffer (containing prote-ase inhibitors and PMSF). 25mL of this solution is added to 2 BigPrep[™] Lysing Matrix B tubes (containing silica beads) and samples are homogenized 4 times for 30 seconds at speed 6.0 m/s. Tubes were incubated on ice for 2 minutes between each run. 80% of yeast cells were lysed.

Is it possible to isolate RNA from paraffin-embedded tissues with the FastPrep[®]-24 System?

Tissue samples to be used in microscopic and histological analyses are often preserved by embedding in paraffin. However, the presence of paraffin may interfere with isolation of RNA. For isolation of RNA from paraffin-embedded tissues, we recommend removing the paraffin by xylene extraction before proceeding with the FastRNA[™] Pro procedure.

Materials:

- Xylene - Ethanol (100%, 95%, 70% solutions)
- Distilled water
- Glass jars or other solvent containers (such as Coplin staining dish)

Procedure:

- 1. Section paraffin blocks at 5-10 microns
- 2. Place sections in a water bath at 42 °C to and eliminate any folds and wrinkles
- 3. Mount sections onto glass slides and let air dry overnight at room temperature. If sections do not adhere to the slides, they can be incubated at 42 °C for up to 8 hours.
- Immerse the slides containing the tissue sections in solvents and solutions as follows:

Xylene 5 minutes Ethanol, 100% 30 seconds Ethanol, 95% 30 seconds Ethanol, 70% 30 seconds, dH₂O 30 seconds

Which settings are recommended for a successful homogenization of skin samples ?

20mg of full thickness skin samples are placed into Lysing Matrix A tubes containing 800µl extraction buffer. Samples are homogenized with the FastPrep[®]-24 instrument, 4 runs of 20 seconds are performed at speed 6.0 m/s. Samples are incubated on ice for 2 minutes between each run.

Are Lysing Matrix tubes resistant to solvents and is it possible to store them at -20°C or -80°C ?

Both tubes and lysing matrix beads are resistant to chemicals (acids, bases, solvents). All Lysing Matrix tubes can be stored in freezers at -20° C and -80° C.

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FastPrep[®]-24 APPLICATIONS

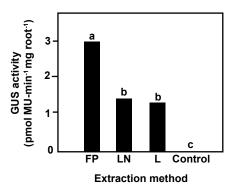


Is the FastPrep[®]-24 System designed for the isolation of enzymes? Is the enzymatic activity preserved?

The FastPrep® system is designed for the isolation of enzymes from any sample. To preserve enzymatic activity, it is recommended to use Lysing Matrix tubes with large beads like 1/4" ceramic beads included in Lysing Matrix A tubes and to homogenize samples for short times with incubation on ice for at least 2 minutes between successive FastPrep® homogenizations in order to prevent overheating of the sample.

Bao J.R. et al. Can.J.Plant Pathol. (2002), Vol 24: 340-348 used the FastPrep® System for the homogenization of tomato roots and isolation of GUS enzyme and demonstrated that this method is yielding the highest enzymatic activity compared to other grinding methods:

Abstract: Insertion of beta-D-glucuronidase (GUS) reporter gene has been found to be useful for detection, quantitation, and monitoring of plant-associated fungi in their environment. GUS was extracted from tomato roots inoculated with a nonpathogenic strain of Fusarium oxysporum, 70T01, that had been genetically modified to express both GUS activity and hygromycin B resistance. To facilitate studies of fungus-plant interactions using the GUS enzyme, we tested several methods for their efficiency of preparation of fungal-encoded GUS from infected plant tissues, namely FastPrep® homogenization, grinding of tissues frozen in liquid nitrogen, and extraction from lyophilized material. Of the three procedures, the Fast-Prep® method yielded the highest GUS activity per unit of inoculated root and provided twice the sensitivity of the other methods. This procedure was also the easiest, quickest, and the most reliable. Up to 12 samples could be analyzed in less than 2 h, and as little as 50 mg of fresh tissue was sufficient. Of the factors examined that could affect extraction efficiency, only the length of homogenization and the presence of protein stabilizers (sucrose, bovine serum albumin, and protease inhibitors) in the GUS buffer improved enzyme activity in the extracts. The FastPrep® method was also highly effective in enumerating fungal colony forming unit (CFU) populations in the root tissues, provided that the timing and speed of homogenization was controlled. Plating of infected root samples homogenized using the FastPrep[®] equipment and a mortar and pestle yielded about 50 times more CFUs per unit root than the colony counts obtained from whole roots, dried and powdered roots, or lyophilized roots.



Comparison of different extraction methods on fungal GUS enzyme activity recovered from tomato roots inoculated with a nonpathogenic strain of Fusarium oxysporum, 70T01, transformed with the GUS gene. Extraction methods included FastPrep® (FP), liquid nitrogen, (LN), and lyophilization and FastPrep® extraction (L). Extracts from noninoculated tomato using the FastPrep® method served as controls. Bars with different letters are significantly different (P < 0.05, Student- Newman-Keuls method, n = 14-15).

Is the FastPrep[®]-24 System designed for DNA extraction from Cryptosporidium oocysts?

There are three main problems associated with the isolation of DNA from Cryptosporidium organisms: (i) the extreme robustness of the oocysts (ii) the different physical and chemical nature of the matrices (faeces, water, food, soil) and their richness in PCR inhibitors (iii) the low number of oocysts usually present in environmental samples.

The reduction or removal of PCR inhibitors is an essential component in the molecular detection of Cryptosporidium in faecal and environmental samples. Currently, pathogen isolation by Immuno Magnetic Separation (IMS) and culture enrichment prior to DNA extraction are standard procedures to eliminate or reduce PCR inhibitors. These methods, however, become impractical for organisms that have no IMS procedures or that cannot be cultured. The use of IMS is also expensive, and this limits the use of samples mostly to single organism detection. Thus, the development of methods for direct extraction of PCR quality DNA is important for the detection of pathogens in environmental samples.

In a recently published study from Jiang J. et al. (Appl. Environ. Microbiol. (2005), Vol 71: 1135-1141), six DNA extraction methods for the detection of Cryptosporidium in water samples were evaluated. The authors concluded that direct DNA extraction with the FastDNA® SPIN kit for soil in combination with the use of a high concentration of BSA represents the most effective tool for PCR detection of Cryptosporidium oocysts in water samples. This reduces the cost of current PCR detection of Cryptosporidium oocysts in water samples significantly as there is no need for the expensive IMS of oocysts prior to DNA extraction. This method also enables the use of extracted DNA for the analysis of other pathogens.

Is it possible to extract DNA from feces with the FastPrep[®]-24 System?

Several studies report the isolation of inhibitor-free DNA from feces with the FastPrep® System for analysis of their bacterial content.

Layton A. et al. Appl.Environ.Microbiol. (2006), Vol 72: 4214-4224 and Ott S.J. et al. J.Clin.Microbiol. (2004), Vol 42: 2566-2572 extracted DNA from human and animal feces with the FastDNA® SPIN Kit for Soil for realtime PCR assays and detection of bacterial species.

On the other side, Tannock G.W. et al. Appl.Environ.Microbiol. (2000), Vol 66: 2578-2588 and Requena T. et al. Appl.Environ.Microbiol. (2002), Vol 68:2420-2427 extracted DNA from 1ml human fecal sample with the FastDNA[™] Kit to monitor the composition of the fecal microflora by PCR-DGGE.

Is there a protocol for RNA extraction from munine corneas?

Corneas are excised from frozen eyes of mice, and RNA is prepared using the FastRNA[™] Pro Green kit. Each cornea is placed in 0.8 mL RNApro[™] solution and homogenized with Lysing Matrix D in a FastPrep® instrument at setting 6.0 for 40 s. After cooling on ice, supernatants are transferred and the lysing matrices rinsed with 0.2 mL RNApro™ solution. Combined supernatants are chloroform-extracted, and RNA is precipitated from the upper phase with an equal volume of isopropanol overnight at -20 °C. Pellets are rinsed with 70% ethanol, air-dried, and resuspended in 10 µL DEPC-treated H₂O at 55-60 °C for 10 min.

Reference: Berglund S.R. et al. (2007) J. Investigative Dermatology Vol 127; 349-353.

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FastPrep[®]-24 APPLICATIONS



Is the FastPrep[®]-24 System suitable for lysis of spores?



Coloured scanning electron micrograph (SEM) of Bacillus anthracis spores

Bacterial and fungal spores either in culture or in environmental samples are successfully lysed with the FastPrep®-24 system. Bacillus subtilis spores in suspension in Lysing Matrix B tubes are processed 3 times for 40 seconds at speed 6.0 m/s with 1 minute cooling on ice between each run. 98% spore lysis was confirmed by microscopy. Hudson K.D. et al. J. Bacteriol. (2005), Vol 183: 4317-4322 Isolated proteins from Bacillus Subtilis spores for Western Blotting. Keijser B.J.F. et al. J. Bacteriol (2007), Vol 189: 3624-3634 purified RNA from cultures of Bacillus Subtilis spores with the FastRNA™ Pro Blue Kit and used total RNA for reverse transcription, labelling and hybridization on micro-array slides.

Anthrax is one of the most dangerous zoonotic infectious disease and has been the first candidate for biological weaponry for over 80 years. It is very difficult to detect anthrax DNA from soil because of the presence of humic acid and many other nonsporulated and sporulated bacteria. DNA was extracted from 1 g of soil artificially contaminated with spores of Bacillus anthracis using a FastDNA[™] SPIN Kit for Soil. Results of nested and realtime PCR experiments indicates that one cell of *B. anthracis* in 1 g of soil is detected by this rapid and highly sensitive method. Cheun H.I. et al. J.Appl. Microbiol. (2005) Vol 95: 728-733.

Two published studies from Roesti D.et al. Appl. Environ. Microbiol (2005), Vol 71: 6673-6679 and Mincer T.J. et al. Appl. Environ. Microbiol (2005), Vol 71: 7019-7028) describe the extraction of DNA from bacterial and fungal spores included in soil cores and marine sediments with the FastDNA® SPIN Kit for Soil. Purified DNA was used for seminested PCR, environmental library construction and DGGE analysis.

Is the FastPrep®-24 System designed for the homogenization of hair samples for forensic toxicology analysis?

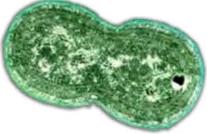
The FastPrep[®]-24 instrument combined with the TeenPrep[™] adapter has been successfully used for the lysis of hair samples prior to drug analysis. Uncut hair samples are added to 15 mL Lysing Matrix tubes containing 8 large 1/4 inch ceramic spheres and 4 steel beads, samples are homogenized 2 times for 50 seconds at speed 6.0 m/s. After the homogenization step, methanol is added to the powder-like homogenate for drug extraction.

What are recommendations for the homogenization of small amounts of animal and plant tissues (less than 10 mg)?

The homogenization of sample amounts lower than 10 mg is performed with Lysing Matrix D tubes after removal of half of the ceramic beads from the tubes. Samples are disrupted at a speed setting of 6.0 m/s for 30 seconds.

Has the FastPrep®-24 System been tested for DNA isolation from blue-green algae?

Blue-green algae also called cyanobacteria are successfully lysed by the FastPrep®-24 Instrument. 2 published studies from Crosbie N.D. et al. Appl. Environ. Microbiol. (2003), Vol 69: 5716-5721 and Steward G.F. et al. App. Environ. Microbiol. (2004), Vol 70: 1455-1465 report the lysis of cyanobacteria cells with the FastPrep® System for DNA extraction experiments. Crosbie N.D. et al. extracted DNA from 10 mL of Synechococcus and Cvanobium cultures using the FastDNA[™] Kit and the FastPrep[®] Instrument. DNA was used for PCR amplification. Steward G.F. et al. added cyanobacteria resuspended in a phenol-chloroform-isoamyl alcohol solution to Lysing Matrix B tubes (0.1 mm silica beads). Samples were homogenized 2 times for 10 seconds at speed 6.0 m/s with the FastPrep® Instrument. Following phase separation and DNA precipitation with ethanol, DNA resuspended in TE buffer was used as template of nested-PCR assays.



Synechococcus cyanobacteria

How to use the FastPrep®-24 System for allergan extractions from air-filter?

A rapid and thorough extraction of endotoxin from PM2.5 air filters has been developed at UCI by employing a high speed shaker (FastPrep®-24, MP Biomedicals, Solon, OH). Briefly, quartz filters were placed into endotoxin-free extraction vials containing pyrogen-free water and processed by the FastPrep® instrument for 30 seconds at 6.5 m/s. Following shaking, the samples were put onto a tube rotator (Dynal Biotech) for 1 hour. An aliquot was assayed using a Limulus Amoebocyte Lysate (LAL) kinetic chromogenic assay (Pyrochrome Associates of Cape Cod, Falmouth, MA). In addition, a negative control filter (blank) was extracted and analyzed. A set of sixteen archived personal PM2.5 air filters were evaluated for the presence of endotoxin. We found detectable endotoxin concentrations in 14 filter extracts with a range of 0.03 - 5.5 EU/mL and a mean concentration of 0.73 EU/mL. The blank filter showed no detectable concentrations of endotoxin.



Quartz filter folded into the 15 mL tube in a TeenPrep™ adapter

Quartz filter lysate after FastPrep[®] lysing with no additional lysing matrix, for 30 seconds at 6.5 m/s speed settings



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