Nucleic acid isolation

cytiva

Unraveling the challenges of nucleic acid isolation

Analyzing nucleic acids is enormously powerful, providing us with insights into a variety of biological processes for basic research and clinical applications. DNA isolation (and RNA isolation) is the first step for many modern genomics techniques and applications, which require high-quality starting material free of contaminants.

How to extract DNA (or RNA)

Genomic DNA extraction is the first step in many molecular biology studies, and all recombinant DNA techniques. Protocols involve breaking open the cells and separating the DNA from other nucleic acids and cellular components in the sample, while also keeping it in good condition for downstream analysis.

The choice of approach depends on several factors, including the target DNA, source organism, the type and quality of your starting material, and the application. They generally all share three common steps: lysis, contaminant removal and DNA recovery.

Step 1: Lysis

Cell lysis involves chemical, mechanical, or enzymatic disruption of cell membranes and denaturation of proteins. The exact method depends on the starting material. Bacteria, mammalian cells, plant cells, and human tissues all might require a slightly different approach.

'Gentle' lysis might involve using a detergent, such as sodium dodecyl sulfate (SDS), or enzymes to break up cell membranes; aggressive lysis might take the form of homogenization to physically break open cell walls.

Step 2: Removing contaminants

You can use both solution-based and solid-phase methods to separate DNA from unwanted lysis debris and potential contaminants. Phenol chloroform DNA extraction, for example, separates water-soluble DNA and denatured proteins into different phases. This is cheap, but slow, and risks carryover of phenol that can affect downstream applications.

Solid-phase extraction binds DNA to a column or bead surface. Silica resins or silica-coated magnetic beads, for example, use chaotropic salts to disrupt hydrogen bonds and bind nucleic acids, enabling contaminants to be washed away. Oligonucleotide-coated resins can also add a level of specificity, but column kits can quickly add up in cost.

Step 3: Recovering the target nucleic acid

Downstream applications require DNA in a suitable format (solvent and concentration). Often, this will be just a matter of precipitating DNA with ethanol, washing, and resuspending in an appropriate buffer. For solid-phase methods, it will first require adjusting the pH or salt concentration of the buffer to release the nucleic acids.

Sample-specific challenges

Cultured mammalian cells and tissues

Cultured cells are relatively easy to lyse with osmotic shock or detergent treatments, while isolating DNA from tissue requires breaking down the extracellular matrix, not just cell membranes. This often requires homogenization followed by silica column (e.g., Amersham™ kits) or mag bead-based (e.g., SeraSil-Mag™) purification, or less favorable phenol-chloroform extraction.

Using formalin-fixed, paraffin-embedded (FFPE) tissue is common in clinical applications and some research studies. It's excellent for preserving tissue structures, but can introduce all sorts of DNA damage with profound effects. That is, as the quality of the DNA isolated directly affects the assay results, positive samples might be overlooked simply because of poor extraction.

Blood

A challenge of DNA extraction from blood is the variability in DNA quantity depending on blood fraction. Red blood cells do not contain DNA, so there's much less per cell in whole blood compared to buffy coat or bone marrow-derived fractions.

Blood coagulation also presents challenges: clotting can prevent effective sample digestion, and some anticoagulants can interfere with PCR amplification.

Bacteria

There are differences between gram-positive and gram-negative samples in DNA extraction from bacteria. Gram-positive samples usually require lysozyme treatment to digest the higher levels of peptidoglycan in the cell wall, whereas for gram-negative samples, a simple osmotic shock might be enough.

DNA is unlikely to be scarce with either type, and it is common to use fast methods, like alkaline extraction and diatomaceous earth, to extract the DNA. Both methods are reliable, but alkaline extraction might not provide the highest purity by itself, and diatomaceous earth can be high cost.

Plant material

Plant cells can be embedded in a tough matrix and have cell walls consisting of glycans and cellulose that are difficult to break. The solvent-based cetyltrimethylammonium bromide (CTAB) extraction method is common for plant material, but it is an aggressive approach. It uses harsh chemicals, is laborious, and often requires further clean-up and optimization for different samples and applications.

Why use magnetic beads for DNA extraction

Magnetic beads provide an excellent alternative to traditional isolation and clean-up methods due to their versatility and ease of use. They do not require additional centrifugation of a potentially already agitated sample, improving the likelihood of recovering larger fragments, and can be scaled up to have a higher binding capacity than columns.

Using magnetic beads is straightforward, needing no hazardous solvents, and releasing the DNA or RNA is just a matter of adjusting the buffer properties (Fig 1). This simplicity also makes magnetic beads well suited to automation in high-throughput applications.

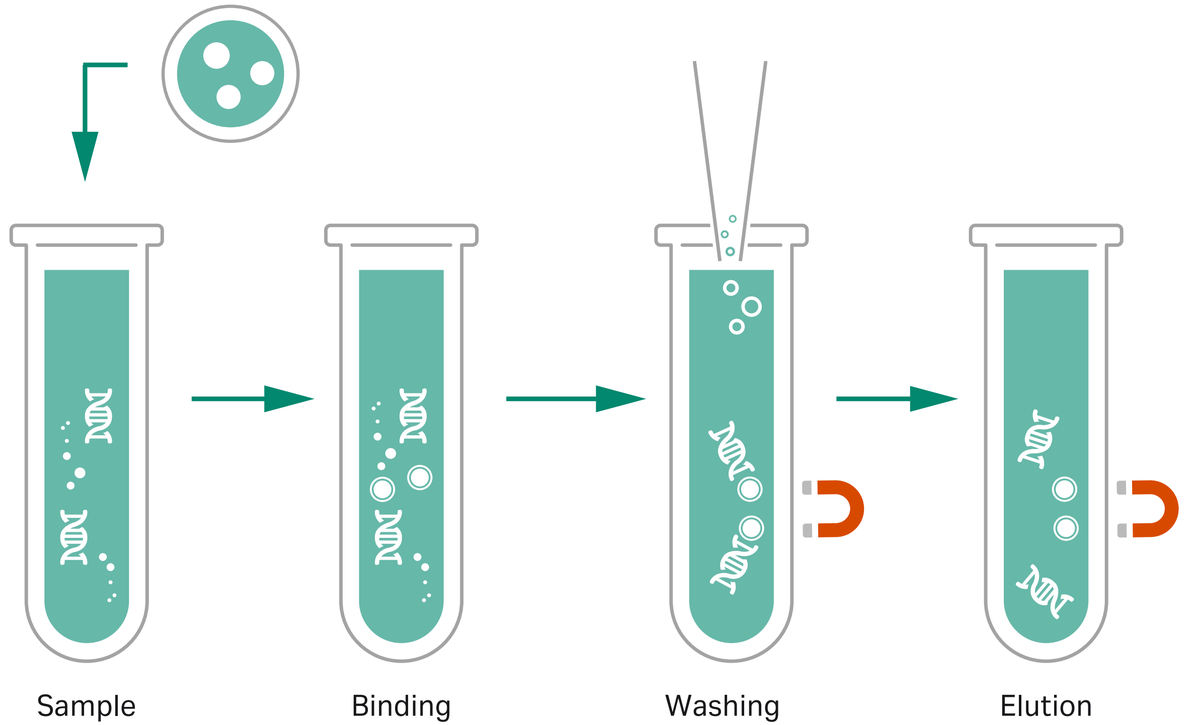


Fig 1. The principle of magnetic beads for nucleic acid isolation.

Nucleon

Nucleon™ systems rapidly extract high molecular weight from whole blood and cultured cells. The Nucleon proprietary resin is added following cell lysis, deproteinization with sodium perchlorate, and a single chloroform extraction.

Nucleon BACC

Nucleon BACC Genomic DNA Extraction Kits are designed for rapid extraction of high-quality, high molecular weight genomic DNA from blood and cell cultures and feature the proprietary Nucleon resin that binds protein while forming a semi-solid stratum during partitioning, which facilitates removal of the aqueous phase and ensures excellent recovery of high quality DNA.

Features and benefits

- Cost effective: Non-column format makes scaling up to large sample volumes easy
- High recovery: Size of recovered DNA ranges from 23 bp to 250 kbp
- Rapid: Phenol-free protocol and only 30 minutes to complete

Product code	Quantity	Description
RPN8501	25	Nucleon BACC1
RPN8512	50	Nucleon BACC3

Find out more

Nucleon HT

Nucleon HT protocols have been designed to prepare high-quality DNA from hard tissues and FFPE tissue sections that is suitable for genomic DNA amplification. Aspiration of the top layer of nucleic acids is simplified and volume of usable sample maximized by a barrier formed between the two layers. The recovered DNA is suitable for a variety of molecular biology applications, including whole genome amplification.

Features and benefits

- Simple: Facilitates the simple, phenol-free extraction of genomic DNA from paraffin-embedded sections and hard tissue requiring proteinase K digestion
- High quality DNA recovery: Each kit is designed to recover high-quality DNA from 50 preparations of up to 25 mg of hard tissue or 50 paraffin sections
- Reproducible: Consistent chloroform extraction of high-quality, amplifiable DNA from formalin-fixed, paraffin-embedded (FFPE) tissue sections

Product code	Quantity	Description
RPN8509	1	Nucleon HT

Find out more

Nucleon PhytoPure

Polysaccharides are common contaminants in plant DNA extracts and can inhibit further enzymatic analysis of DNA. Nucleon PhytoPure DNA extraction system has been developed specifically to solve this problem and has been used successfully on a wide range of fresh, frozen or freeze-dried plant material.

Features and benefits

- Fast: Enables extraction of DNA in less than 1 hour
- Reliable: Eliminates the need to use phenol
- Simple: Easy-to-use protocol requires only one centrifugation step prior to DNA precipitation
- Pure: DNA is of high quality and suitable for RFLP, RAPD and AFLP analyses

Product code	Quantity	Description
RPN8510	50 preparations of 0.1 g	Nucleon PhytoPure
RPN8511	50 preparations of 1.0 g	Nucleon PhytoPure

Find out more

GenomicPrep Mini Kits

Find out more

The genomicPrep Mini Kits deliver high quality intact high molecular weight gDNA from a variety of samples using a convenient and efficient protocol. The kit delivers high yields of highly pure gDNA suitable for use in a variety of downstream molecular biology workflows including enzyme digests, cloning, electrophoresis, qPCR, genotyping and sequencing.

Blood genomicPrep Mini Spin Kits

Blood genomicPrep Mini Spin Kit is designed for the rapid extraction and purification of high molecular weight genomic DNA (gDNA) from whole blood, buffy coat, bone marrow, and nucleated red blood cells. Uses chaotropic agents to extract DNA from blood cells, denature protein components, and promote the selective binding of DNA to a column-based, novel silica membrane.

Features and benefits

- Fast results: Streamlined workflow reduces the number of pipetting volume changes and the overall number of steps to deliver sample to gDNA results in 15 minutes
- Minimal shearing: Resulting in the production of 5 to 10 µg of good quality, intact genomic DNA from a 200 µL sample. One kit handles a wide range of blood sample types and volumes from 50 µL to 1 mL.
- High quality: Gentle room temperature lysis conditions deliver high-quality, > 97% intact DNA with an average size of > 20 kb

Product code	Quantity	Description
28904263	10	Blood genomicPrep Mini Spin Kit
28904264	50	Blood genomicPrep Mini Spin Kit
28904265	250	Blood genomicPrep Mini Spin Kit

Amersham Tissue and Cells genomicPrep Mini Spin Kits

Genomic DNA purifications performed with the Amersham Tissue and Cells genomicPrep Mini Spin Kit yield consistent results and are highly robust across different sample types.

Features and benefits

- Fast results: Reduces time from tissue sample to gDNA and produces high-quality product in just 90 minutes
- Simpler purification: Color-coded caps and bottles with matching protocol steps minimize the chance for error
- High quality and purity: Optimized tissue protocol produces intact, RNA-free gDNA that is > 20 kb in size with a purity of 1.8 (A_{260}/A_{280})

Product code	Quantity	Description
28904274	10	Amersham Tissue and Cells genomicPrep Mini Spin Kit
28904275	50	Amersham Tissue and Cells genomicPrep Mini Spin Kit
28904276	250	Amersham Tissue and Cells genomicPrep Mini Spin Kit

Find out more

Amersham Bacteria genomicPrep Mini Spin Kits

Amersham Bacteria genomicPrep Mini Spin Kit is designed for the rapid extraction and purification of high molecular weight genomic DNA (gDNA) from Gram-negative (G-ve) and Gram-positive (G+ve) bacteria. The procedure for G-ve bacteria can be completed in about 40 minutes (sample to gDNA).

Features and benefits

- Fast results: Streamlined workflow reduces the number of pipetting volume changes and the overall number of steps
- Optimized kit: Dedicated kit optimized for bacterial gDNA with separate protocols for G-ve and G+ve bacteria
- Ease of use: Color-coded caps and bottles with matching protocol steps minimize the chance for error
- High quality and purity: Optimized protocol produces intact, RNA-free gDNA that is > 20 kb in size with a purity > 1.8 (A_{260}/A_{280})

Product code	Quantity	Description
28904257	10	Amersham Bacteria genomicPrep Mini Spin Kit
28904258	50	Amersham Bacteria genomicPrep Mini Spin Kit
28904259	250	Amersham Bacteria genomicPrep Mini Spin Kit

Find out more

Amersham plasmidPrep Mini Spin Kit

Amersham plasmidPrep Mini Spin Kit is designed for the rapid extraction and purification of plasmid DNA. The procedure can be completed in less than 10 minutes to yield plasmid DNA with a purity and quality suitable for molecular biology applications including sequencing, PCR, and cloning.

Amersham plasmidPrep Mini Spin Kit uses a simple plasmid DNA purification protocol involving a modified alkaline lysis procedure and a novel silica-based membrane to achieve highly efficient plasmid DNA purification. The typical plasmid DNA yield from a fresh 1.5 mL culture of *E. coli* containing a high copy number plasmid is approximately 9 µg.

Instead of organic solvents, the kit contains chaotropic salts to denature protein components and promote selective binding of DNA to the novel silica membrane. The kit contains spin columns prepacked with a novel silica membrane, suspension, lysis, and neutralization buffers containing chaotropic salts, wash and elution buffers, microcentrifuge collection tubes, a full protocol booklet, and a detachable, quick reference protocol card.

Features and benefits

- Save time: < 10 min preparation, a 50% reduction in protocol time from QIAprep™ Spin Miniprep Kit
- Easy to follow: quick reference protocol card provides instructions at a glance for experienced users
- Simple storage: Amersham buffers are stored at room temperature keeping the kit intact
- High purity: plasmid DNA compatible with salt-sensitive enzymes such as HindIII; minimal particulate matter (as measured by A_{340}/A_{260})
- High quality: Phred 20 scores (ANOVA p-value > 0.05) and amount of supercoiled plasmid DNA equivalent to that of QIAprep Spin Miniprep Kit

Find out more

Product code	Quantity	Description
28904271	10	Amersham plasmid Mini Spin (By Request)
28904269	50	Amersham plasmidPrep Mini Spin Kit
28904270	250	Amersham plasmidPrep Mini Spin Kit

Amersham plasmidPrep Midi Flow Kit

Find out more

Amersham plasmidPrep Midi Flow Kit is designed for the purification of high yields of transfection-grade plasmid DNA. The procedure utilizes the superior capacity and selectivity of the Fast Flow plasmid purification medium to facilitate processing of larger culture volumes providing greater overall yields of plasmid DNA with low levels of endotoxin contamination.

Purified plasmid DNA is suitable for use in a range of demanding cellular and molecular biological applications including: transfection, enzymatic modification and amplification, as well as automated sequencing.

Features and benefits

- Better performance: up to seven times lower endotoxin levels in a side-by-side comparison with Qiagen™, resulting in increased plasmid DNA purity
- Increased yield: plasmidPrep midi column can accommodate twice the culture volume of Qiagen Plasmid Midi Kit, resulting in yields in excess of 300 µg
- Convenience: columns are pre-equilibrated and ready to use
- Easy to use: our Fast Flow media reduces the risk of clogging, allowing twice the input material in comparison with Qiagen*, so more sample can be processed faster than existing methods
- User friendly: color-coded caps and bottles with matching protocol steps minimize the chance for error
- Easy to follow: quick reference protocol card provides instructions at a glance for experienced users

* Compared with Qiagen Plasmid Midi Kit

Product code	Quantity	Description
28904267	25	Amersham plasmidPrep Midi Flow Kit
28904268	100	Amersham plasmidPrep Midi Flow Kit

Amersham RNAspin Kits

Our Amersham RNA purification kits are designed to ensure that you get the reproducibility, yield and purity you need, with minimal degradation in every experiment. Amersham kits accommodate a diverse range of application requirements and sample types, delivering RNA that can be used for downstream applications, such as qRT-PCR and microarray analysis.

Amersham RNAspin Mini Kits

By identifying key elements that affect the quality of preparation the RNAspin Mini Kit allows total RNA isolation from diverse sample types, resulting in the extraction of RNA suitable for use in sensitive downstream applications such as microarray analysis and quantitative or endpoint RT-PCR. Our on-column DNase I digest improves purities by addressing issues of gDNA contamination.

Features and benefits

- Maximized yields: the inclusion of prefilters and a unique lysis buffer makes it less susceptible to foaming
- Efficient: Column-binding capacity of 100 µg and elution volumes as low as 40 µL eliminate the need to concentrate sample
- Reliable: Well-established silica-membrane technology

Product code	Quantity	Description
25050087	10	Amersham RNAspin Mini Kit
25050070	20	Amersham RNAspin Mini Kit
25050071	50	Amersham RNAspin Mini Kit
25050072	250	Amersham RNAspin Mini Kit

Find out more

Amersham RNAspin 96 Kit

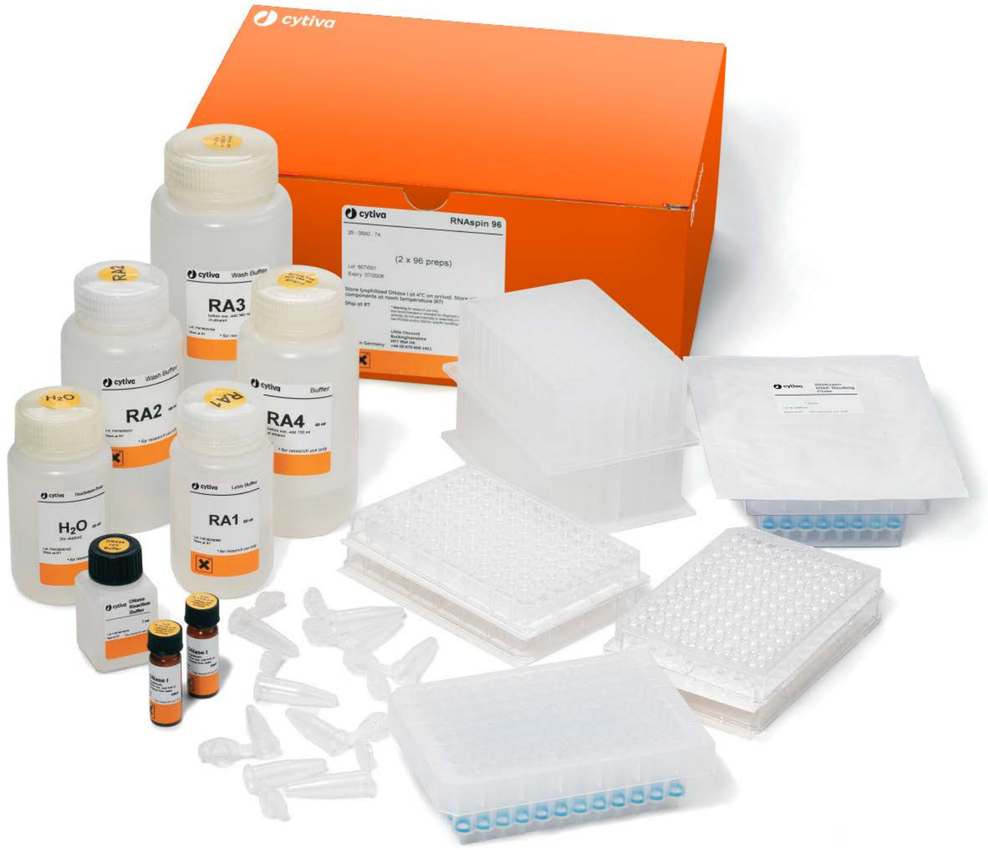
With protocol run times that have been optimized to be as short as possible, Amersham RNAspin 96 kits support a high throughput approach, whether samples are processed under vacuum, using centrifugation, manually, or with automation. The on-column lysis for small amounts of sample improves efficiency by avoiding mechanical homogenization.

Features and benefits

- Fast and efficient: Purification in a 96-well format with high reproducibility in less than 70 minutes
- Supports automation: Integrated wash plate eliminates risk of cross contamination and compatible with common liquid handling instruments for fast integration
- Convenient: DNase I included for convenient on-column gDNA removal, leading to pure total RNA
- Flexible and scalable: Includes prefilter accessory plate

Product code	Quantity	Description
25050075	4 × 96 preps	Amersham RNAspin 96 Kit

Find out more



TriplePrep Kit

The Kit is designed for rapid, simultaneous extraction and isolation of high-quality genomic DNA (gDNA), total RNA, and total denatured proteins from animal tissues and mammalian cells. High yields of high-quality DNA, RNA, and proteins can be extracted in less than 1 h using a flexible, easy-to-follow workflow allowing researchers to directly correlate data generated from the same sample.

Fast and simple — the Kit is designed to go from sample to DNA/RNA/protein in less than 1 h — streamlined workflow reduces the overall number of steps, resulting in up to 70% time saving compared to preparing each analyte. Easy to follow — color-coded caps and bottles with matching protocol steps minimize the chance for error. A quick-reference protocol card provides instructions at a glance for experienced users.

Features and benefits

- High yield: High DNA, RNA, and protein yields from small samples — optimized buffer, columns, and protocol ensure high recovery of gDNA, total RNA, and total denatured proteins
- High quality: High purity — DNA-free RNA, with DNase provided in the kit. Suitable for downstream applications — gDNA, RNA, and proteins validated by numerous downstream applications.
- Easy to use: Flexible workflow — can isolate any two or all three analytes with multiple stop points in the protocol. User friendly — minimal change of centrifugation speed, time, and pipetting volume.

Product code	Quantity	Description
28942544	50	Prep Kit

Find out more

Sera-Mag Oligo (dT) Coated Magnetic particles

Colloidally stable Sera-Mag™ Oligo (dT) magnetic particles contain covalently bound oligo (dT)14 and will remain in suspension for extended periods of time in the absence of a magnetic field, making them well suited for capturing or isolating mRNA from a variety of sources.

Oligo (dT) particles can also be used as a universal base particle for coupling unique oligo sequences. Simply synthesize the oligo with a poly-A tail for easy attachment to the oligo (dT) particles.

Features and benefits

- Versatile: Once isolated, selective purification of mRNA from total RNA for NGS, RT-PCR, cDNA library construction, or subtractive hybridization can be performed
- Performance: The approximate mRNA binding-capacity is 11 µg of mRNA per mg of particles (dependent upon sample and message length)

Product code	Quantity	Description
38152103011150	1 mL	Sera-Mag Oligo (dT) Coated Magnetic particles
38152103010150	5 mL	Sera-Mag Oligo (dT) Coated Magnetic particles
38152103010350	100 mL	Sera-Mag Oligo (dT) Coated Magnetic particles

Find out more

Sera-Mag Carboxylate and SpeedBead Carboxylate

Carboxylic groups on the surface of Sera-Mag SpeedBeads and Sera-Mag Carboxylate-Modified Magnetic Beads permit easy covalent coupling to target biomolecules of interest, such as proteins and nucleic acids, using convenient carbodiimide chemistry.

The cauliflower-shaped surface paired with proprietary Sera-Mag and SpeedBead chemistry, provides a large surface area and offers excellent sensitivity and low non-specific binding for greater accuracy. This can maximise sample retention or reduce the amount of beads required.

The beads are available with different levels of hydrophobicity/ hydrophilicity and magnetite layering.

Find out more

Sera-Mag Carboxylate-Modified Magnetic Particles

Sera-Mag Carboxylate-Modified Magnetic Beads combine a fast magnetic response time and high binding capacity, sensitivity, stability and physical integrity.

Features and benefits

- Ease of use: Covalent coupling of proteins, nucleic acids, etc. to carboxyl groups on the surface using standard coupling technologies
- Convenient: Isolation, selection and clean-up of nucleic acids or direct conjugation of specific oligos and enzymes

Product code	Quantity	Description
24152105050250	15 mL	Sera-Mag Carboxylate-Modified Magnetic Particles (Hydrophylic)
24152105050350	100 mL	Sera-Mag Carboxylate-Modified Magnetic Particles (Hydrophylic)
24152105050450	1000 mL	Sera-Mag Carboxylate-Modified Magnetic Particles (Hydrophylic)
44152105050250	15 mL	Sera-Mag Carboxylate-Modified Magnetic Particles (Hydrophobic)
44152105050350	100 mL	Sera-Mag Carboxylate-Modified Magnetic Particles (Hydrophobic)
44152105050450	1000 mL	Sera-Mag Carboxylate-Modified Magnetic Particles (Hydrophobic)

Find out more

Sera-Mag SpeedBead Carboxylate-Modified Magnetic Particles

Sera-Mag Speedbeads have a second layer of magnetite applied through the same core shell design process, allowing a reaction twice as fast as the Sera-Mag Carboxylate-Modified beads when in the presence of a magnetic field. Speedbeads are especially useful where the reaction medium is highly viscous, and in clinical assays requiring a faster magnetic response time.

Features and benefits

- Convenient: Isolation, selection and clean-up of nucleic acids or direct conjugation of specific oligos and enzymes
- Reliable: Fast, precise and high binding capacity for sample preparation, nucleic acid isolation, proteomics and immunoassay applications

Product code	Quantity	Description
45152105050250	15 mL	Sera-Mag SpeedBead Carboxylate (Hydrophylic)
45152105050350	100 mL	Sera-Mag SpeedBead Carboxylate (Hydrophylic)
65152105050250	15 mL	Sera-Mag SpeedBead Carboxylate (Hydrophobic)
65152105050350	100 mL	Sera-Mag SpeedBead Carboxylate (Hydrophobic)
65152105050450	1000 mL	Sera-Mag SpeedBead Carboxylate (Hydrophobic)

Find out more

SeraSil-Mag Magnetic Particles

SeraSil-Mag silica coated superparamagnetic beads deliver a high purity extraction solution for highly sensitive applications when sample is scarce. The beads provide an optimal binding surface, with regular morphology, to optimize binding efficiency and reduce variability, simplifying the transition from column purification to bead-based purification.

Find out more

Features and benefits

- High iron oxide content (60 emu/g): Fast magnetic response (~ 5 s) shortens time of magnetic steps during isolation
- Uniformity: Particles are uniform in size (submicroscale diameter 700 nm and 400 nm [monodispersed]), providing narrow size distribution
- Low sedimentation rate: Good buoyancy enhances ease of handling, automation, and reproducibility
- Purity: Used to isolate and purify genomic DNA from whole human blood providing A_{260}/A_{280} ratios between 1.70–1.90 and A_{260}/A_{230} ratios as high as 2

Product code	Quantity	Description
29357369	5 mL	SeraSil-Mag 400
29357371	60 mL	SeraSil-Mag 400
29357372	450 mL	SeraSil-Mag 400
29357373	5 mL	SeraSil-Mag 700
29357374	60 mL	SeraSil-Mag 700
29357375	450 mL	SeraSil-Mag 700

cytiva.com/genomics-diagnostics

For local office contact information, visit cytiva.com/contact

CY17355-17Mar21-HB

cytiva