Product Information Sheet # SCO500, # SCO510







Product

MobiSpin G-Columns pre-packed with Sephadex[®] G-50 resin and equilibrated in STE buffer (10mM Tris/HCl pH 8.0, 100 mM NaCl, 1 mM EDTA).

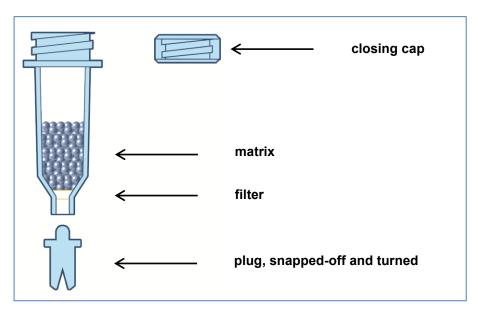


Fig.1: MobiSpin G-Column

The MobiSpin G-Column is closed with a screw cap and contains the Sephadex[®] G-50 matrix equilibrated with STE buffer. The tip of the MobiSpin Column is a fixed snap-off plug. Prior to the first centrifugation step it has to be bent down and removed. The plug can be reused for closing by turning it upside down.

Description

The MobiSpin G-Columns are prepacked with Sephadex G-50 resin, equilibrated and ready to use. The Sephadex G-50 matrix is a hydrophilic matrix that swells in buffer and acquires its chromatographic properties under wet conditions. In this state beads have a pore size of 700 Da that allow, e.g., hydrated salt ions or dNTPs (dye-labeled or radiolabeled) to enter into the pores while DNA/RNA > 20 bases and most other biomolecules stay outside.

Sephadex® is a registrered trademark of GE Healthcare

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Features

- Compatible with laboratory standard
- Easy handling: spin, load sample, spin and collect the purified product
- Columns are prepacked, equilibrated, and ready to use
- Very fast procedure: sample purification in less than 4 minutes
- · Reproducible results with simplified protocols
- Numerous samples can be processed simultaneously
- No sample dilution
- Large number of applications concerning nucleic acid purification:
 - desalting of nucleic acids,
 - buffer exchange, and
 - removal of dye terminators or unincorporated labeled nucleotides from DNA labeling reactions.

Applications

Desalting and Buffer exchange

MobiSpin G-Columns pre-packed with G-50 Sephadex[®] resin remove small molecules like salt ions from DNA/RNA (even from dsDNA fragments as small as 20 bp) and are therefore most suitable for buffer exchange.

Removal of dye terminators or unincorporated labeled nucleotides

MobiSpin G-Columns pre-packed with Sephadex [®] G-50 resin are particularly suited for the removal of fluorescent dye dideoxyterminators (e.g., Cy5/Cy3 nucleotides) from cycle sequencing reactions.

Furthermore, these columns are convenient for the extraction of unincorporated labeled nucleotides (dyelabeled or radiolabeled dNTPs or ddNTPs) from DNA labeling reactions, e.g., PCR probe labeling, Nick Translation, or DNA end-labeling. The purified DNA is applicable to downstream applications like FISH (fluorescence *in situ* hybridization) or Southern/Northern blotting. The removal of unincorporated labeled nucleotides is a precondition for determining the DNA labeling rate. For good recovery rates, labeled DNA fragments must be at least 20 bp in length.

Protocol

- · Resuspend the resin in the column by vortexing.
- Bend off the tip of the column and loose the cap one fourth turn.
- Place the column in a 1.5 ml microcentrifuge tube and use this as collecting vessel.
- Pre-spin the column 1 minute at 735 x g in a microcentrifuge with a fixed-angel rotor. Do not pulse as this will override the variable speed setting. Please consider centrifugation note below!
- Use the column **immediately** after removing the equilibration buffer from the resin to avoid drying up!

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Note: before using a MobiSpin G-Column, it may be of significant importance to calculate the speed at which the column should be centrifuged. The calculation of the appropriate centrifugation speed ensues from the following formula:

RCF =
$$(1.12) \times (r) \times (rpm/1000)^2$$
,

whereby RCF is the relative centrifugal force, r the radius measured in mm from the center of the spindle to bottom of rotor bucket, and rpm the revolutions per minute.

Example:

for a force of 735 g the above equation resolves to

$$rpm = 1000 \sqrt{657/r}$$

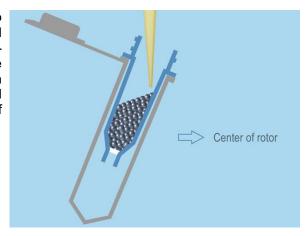
r = radius in mm measured from center of spindle to bottom of rotor bucket

rpm = revolutions per minute

With a rotor having a radius of 110 mm, the appropriate speed would be 2444 rpm.

- Place the column in a new 1.5 ml tube and slowly apply the sample (50 μl or less) to the upper side of the slanted matrix surface as shown in Fig. 2. Take care not to disturb the resin bed!
- Spin the column 2 minutes at 735 x g. The purified sample is collected in the bottom of the support tube.

Fig. 2: Load the sample onto the higher side of the slanted matrix surface. For subsequent centrifugation the column should be placed with the higher side of the slanted matrix toward the center of the rotor.



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Order Information, Shipping, and Storage

Order#	Product	Amount
SCO500	MobiSpin G-50	20 columns
SCO510	MobiSpin G-50	100 columns
shipped at RT; store at 4°C		

Related Products

Order#	Product	Amount
SCO100	Desalting MobiSpin Columns	20 columns
SCO110	Desalting MobiSpin Columns	100 columns
SCO200	MobiSpin S-200	20 columns
SCO210	MobiSpin S-200	100 columns
SCO300	MobiSpin S-300	20 columns
SCO310	MobiSpin S-300	100 columns
SCO400	MobiSpin S-400	20 columns
SCO410	MobiSpin S-400	100 columns
SCO234	MobiSpin S-200, S-300, S-400	3 x 10 columns

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