

Micro Bio-Spin® 30 Columns in RNase-Free Tris

Introduction

Micro Bio-Spin columns are designed for rapid cleanup and purification of riboprobes, including removal of unincorporated nucleotides or templates used for in vitro transcription. The columns are packed with a special grade of Bio-Gel P-30 polyacrylamide gel matrix manufactured by Bio-Rad specifically for spin columns. This unique gel produces very efficient, noninteractive separation by size. Micro Bio-Spin columns are suitable for use with 1.5 or 2.0 ml microcentrifuge tubes and are autoclavable.

Quality Control

Each lot of Micro Bio-Spin 30 RNase-free columns is tested and certified RNase free. The QC procedure for the detection of RNase activity involves purification of in vitro synthesized transcripts. The purified transcripts are incubated at room temperature for 2 hr. The quality of the transcript is checked by agarose gel electrophoresis.

Technical Information

Gel Matrix

The columns are packed with Bio-Gel P-30 polyacrylamide gel suspended in 1 ml Tris buffer to heading level (10 mM Tris-HCl, pH 7.4) with 0.02% sodium azide.

Sample Volume

The column accommodates sample loads of 10–75 µl. Volumes <10 µl or >75 µl may affect column performance.

Exclusion Limits

Micro Bio-Spin 30 columns are used to purify nucleic acids larger than 20 bases or bp. Recovery of fragments smaller than 20 bases will be lower.

Centrifuge Type

Centrifuge in a microcentrifuge at 1,000 x g.

Autoclavability

Micro Bio-Spin 30 columns and wash and collection tubes are completely autoclavable. If a sterile Micro Bio-Spin column is required, autoclave the column at 121°C for 20–30 min on the liquid cycle. If exchanging buffers, the buffer pH in the column should be in the range 6.0–8.0 prior to autoclaving.

Chemical Stability

The columns are stable at pH 2–10 in common aqueous buffers, formamide, dilute organic acids, alcohol, 20%(v/v) other chaotropic agents, and detergents.

Storage

Store at 4°C. Do not freeze. The shelf life is 1 year at 4°C.

Instructions for Use

1. Invert the column sharply several times to resuspend the settled gel. Tap the column to remove all air bubbles. Snap off the tip and place the column in a 2.0 ml microcentrifuge tube (supplied). Remove the cap. If buffer does not begin to flow from the column, push the cap back on the column and remove it again to start the flow. Allow the excess packing buffer to drain by gravity to the top of the gel bed (about 2 min). Discard the drained buffer and place the column back into the 2.0 ml tube.
2. Centrifuge for 2 min in a microcentrifuge at 1,000 x g (see Centrifugation Notes section) to remove the remaining packing buffer. Note: The speed is important to ensure proper performance of the columns. Discard the tube.
3. Place the column in a clean 1.5 ml microcentrifuge tube (supplied with 25 and 100 packs). Carefully apply the sample (10–75 µl) directly onto the top center of the gel bed. Do not disturb the gel bed. Application of more or less than the recommended sample volume may decrease column performance.
4. After loading the sample, centrifuge the column for 4 min at 1,000 x g.
5. The purified sample is now in 10 mM Tris buffer.
6. Properly dispose of the used column.

Buffer Exchange

The gel in the Micro Bio-Spin columns is suspended in 10 mM Tris buffer, pH 7.4. The gel matrix is compatible with most aqueous buffers. Buffer exchange can be achieved using the following procedure.

1. Follow steps 1 and 2 in the Instructions for Use section.

- Apply the new buffer in 500 µl aliquots. After each application of new buffer, let the buffer drain out by gravity, or centrifuge the column for 1 minute at 1,000 x g to remove the buffer. Discard buffer from the wash tube. Repeat as required. Three washes result in > 99% buffer exchange. Four washes result in > 99.9% buffer exchange.
- Sample can now be applied to the column as directed in steps 3 through 6 in the Instructions for Use section.

Centrifugation Notes

Micro Bio-Spin columns fit in 1.5 ml microcentrifuge tubes for sample collection during centrifugation. Use the 2 ml wash tubes provided with the columns for the initial column equilibration step.

Benchtop microcentrifuges capable of generating a minimum force of 1,000 x g are suitable for Micro Bio-Spin column use. The gravitational force created at a particular speed is a function of the radius of the microcentrifuge rotor. Consult the micro-centrifuge instruction manual for conversion information from rpm to g-force. Alternatively, to calculate the rotational speed required to reach a gravitational force of 1,000 x g, use the following equation:

$$g = (1.12 \times 10^{-5})v^2r$$

where r is the radius in centimeters measured from the center of the rotor to the middle of the Micro Bio-Spin column, and v is the speed of the rotor in rpm.

Ordering Information

Catalog #	Description
RNase-Free Micro Bio-Spin Columns With Bio-Gel P-30 in Tris Buffer (Blue Cap)	
732-6250	Micro Bio-Spin 30 Columns, RNase free, 25
732-6251	Micro Bio-Spin 30 Columns, RNase free, 100

Micro Bio-Spin Columns With Bio-Gel P-30 in Tris Buffer (Orange Cap)	
732-6223	Micro Bio-Spin 30 Columns, 25
732-6224	Micro Bio-Spin 30 Columns, 100
732-6226	Micro Bio-Spin 30 Columns, 1,000

Micro Bio-Spin Columns With Bio-Gel P-6 in Tris Buffer (Green Cap)	
732-6221	Micro Bio-Spin 6 Columns, 25
732-6222	Micro Bio-Spin 6 Columns, 100
732-6225	Micro Bio-Spin 6 Columns, 1,000

Catalog #	Description
Micro Bio-Spin Columns With Bio-Gel P-30 in SSC Buffer (Yellow Cap)	
732-6202	Micro Bio-Spin 30 Columns, 25
732-6203	Micro Bio-Spin 30 Columns, 100
732-6206	Micro Bio-Spin 30 Columns, 1,000
Micro Bio-Spin Columns With Bio-Gel P-6 in SSC Buffer (Clear Cap)	
732-6200	Micro Bio-Spin 6 Columns, 25
732-6201	Micro Bio-Spin 6 Columns, 100
732-6205	Micro Bio-Spin 6 Columns, 1,000
Empty Columns	
732-6204	Empty Micro Bio-Spin Columns, 100
731-1660	End Caps for Micro Bio-Spin Columns, 1,000
SEQueaky Kleen™ H₂O Dye Terminator Removal Kit	
732-6500	SEQueaky Kleen H ₂ O Dye Terminator Removal Kit, 2 x 96-well
732-6505	SEQueaky Kleen H ₂ O Bulk Kit, 50 x 96-well
PCR Kleen™ Spin Columns (Red Cap)	
732-6300	PCR Kleen Spin Columns, 25
732-6301	PCR Kleen Spin Columns, 100
Freeze 'N Squeeze™ DNA Gel Extraction Kit	
732-6165	Freeze 'N Squeeze Spin Columns, 25
732-6166	Freeze 'N Squeeze Spin Columns, 100



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