



Micro Bio-Spin® Chromatography Columns

Introduction

Micro Bio-Spin chromatography columns are ready to use for rapid and efficient cleanup and purification of nucleic acids and proteins using a microcentrifuge.

Micro Bio-Spin Columns

- Remove dye terminators
- · Remove unincorporated nucleotides
- · Desalt nucleic acids, proteins and peptides

The columns are packed with special grades of Bio-Gel® P polyacrylamide P-6 or P-30 gel matrices manufactured specifically for Bio-Rad spin columns. This unique gel produces very efficient, non-interactive size separations. Micro Bio-Spin columns are suitable for use with 1.5 or 2.0 ml microcentrifuge tubes and are completely autoclavable.

Technical Information

Gel Matrix

Bio-Gel P-6 or P-30 polyacrylamide gel suspended in $1.0~\mathrm{ml}$ of buffer

Buffers

SSC buffer (150 mM sodium chloride, 17.5 mM sodium citrate, pH 7.0) with 0.02% sodium azide

Tris buffer (10 mM Tris-HCl, pH 7.4) with 0.02% sodium azide

Sample Application Volumes

Nucleic acids, proteins, and peptides, 20–75 $\mu l.$ Volumes less than 20 μl may affect recovery

Exclusion Limits

Bio-Gel P-6 gel: 5 base pairs (nucleic acids) or 6,000 daltons (proteins, peptides)

Bio-Gel P-30 gel: 20 base pairs (nucleic acids) or 40,000 daltons (proteins, peptides)

Expected Retention and Recovery

Micro Bio-Spin 6 column

98% retention of unincorporated nucleotides at 20 μ l 90% recovery of applied DNA at 20 μ l

Micro Bio-Spin 30 column

100% retention of unincorporated nucleotides at 20 μl 95% recovery of applied DNA at 70 μl

Centrifuge Type

Microcentrifuge with a centrifugal force of 1,000 x (g).

Autoclavability

Micro Bio-Spin columns, Bio-Gel P gel, and collection tubes are completely autoclavable at 121 °C for 30 minutes at pH 6.0–8.0.

Chemical Stability

pH 2–10, common aqueous buffers, formamide, dilute organic acids, alcohol, 20% (V/V) other chaotropic agents, detergents.

Storage

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

Instructions for Use

- 1. Invert the column sharply several times to resuspend the settled gel and remove any bubbles. Snap off the tip and place the column in a 2.0 ml microcentrifuge tube (included). Now remove the top cap. If the column does not begin to flow, push the cap back on the column and then remove it again to start the flow. Allow the excess packing buffer to drain by gravity to the top of the gel bed (about two minutes). Discard the drained buffer then place the column back into the 2.0 ml tube.
- 2. Centrifuge for 2 minutes in a microcentrifuge at 1,000 x (g) (see Centrifugation Notes section) to remove the remaining packing buffer. Discard the buffer.
- 3. Place the column in a clean 1.5 or 2.0 ml microcentrifuge tube. Carefully apply the sample (20–75 μ l) directly to the center of the column. Application of more or less than the recommended sample volume may decrease column performance.
- 4. After loading sample, centrifuge the column for 4 minutes at $1,000 \times (g)$.
- 5. Following centrifugation, the purified sample is now in either SSC or Tris buffer. Molecules smaller than the column's exclusion limit will be retained by the column (see Specifications).
- 6. Properly dispose of the used column.

Buffer Exchange

The gel in the Micro Bio-Spin columns is suspended in either SSC buffer, pH 7.0, or Tris buffer, pH 7.4. The gel matrix is compatible with most aqueous buffers. Buffer exchange can be achieved using the following procedure.

- 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 to remove the buffer. Discard buffer from collection tube. Repeat as required. Three washes result in > 99% of the buffer exchanged. Four washes result in > 99.9% of buffer exchanged.
- 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 1.5 ml microcentrifuge tubes for sample collection during centrifugation. Use the 2.0 ml microtubes 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 rpm is a function of the radius of the microcentrifuge rotor. Consult the microcentrifuge instruction manual for conversion information from rpm to g-force. Alternatively, to calculate the speed (rpm) required to reach a gravitational force of 1,000 x (g), use the following equation:

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

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

Sample Dilution Troubleshooting Guide

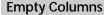
Sample dilution will occur if, during column equilibration, the column is centrifuged without first removing the excess packing buffer. Dilution will also occur if the excess packing buffer is left in the collection tube prior to centrifugation. The tip of the column becomes submerged and retains excess buffer.

To correct this situation, always discard the buffer from the collection tube after gravity draining and after the first centrifugation (steps 1 and 2 of Instructions for Use section).

Sterilization

If a sterile Micro Bio-Spin column is required, autoclave the column at 121 °C for 20–30 minutes. If exchanging buffers, the buffer pH in the column should be in the range of 6.0 to 8.0 prior to autoclaving.

Ordering Information		
Catalog Number	Product Description	
Bio-Gel P-6 Columns in SSC		
732-6200	Micro Bio-Spin 6 Column, SSC, 25, includes 25 P-6 columns in SSC buffer, 25 buffer collection tubes, and instruction manual	
732-6201	Micro Bio-Spin 6 Column, SSC, 100, includes 100 P-6 columns in SSC buffer, 100 buffer col- lection tubes, and instruction manual	
732-6205	Micro Bio-Spin 6 Column, SSC, 1,000, includes 1,000 P-6 columns in SSC buffer, 1,000 buffer collection tubes, and instruction manual	
Bio-Gel P-6 Columns in Tris		
732-6221	Micro Bio-Spin 6 Column, Tris, 25, includes 25 P-6 columns in Tris buffer, 25 buffer collection tubes, and instruction manual	
732-6222	Micro Bio-Spin 6 Column, Tris, 100, includes 100 P-6 columns in Tris buffer, 100 buffer col- lection tubes, and instruction manual	
732-6225	Micro Bio-Spin 6 Column, Tris, 1,000, includes 1,000 P-6 columns in Tris buffer, 1,000 buffer collection tubes, and instruction manual	
Bio-Gel P-3	30 Columns in SSC	
732-6202	Micro Bio-Spin 30 Column, SSC, 25, includes 25 P-30 columns in SSC buffer, 25 buffer collection tubes, and instruction manual	
732-6203	Micro Bio-Spin 30 Column, SSC, 100, includes 100 P-30 columns in SSC buffer, 100 buffer collection tubes, and instruction manual	
732-6206	Micro Bio-Spin 30 Column, SSC, 1,000, includes 1,000 P-30 columns in SSC buffer, 1,000 buffer collection tubes, and instruction manual	
Bio-Gel P-3	30 Columns in Tris	
732-6223	Micro Bio-Spin 30 Column, Tris, 25, includes 25 P-30 columns in Tris buffer, 25 buffer collection tubes, and instruction manual	
732-6224	Micro Bio-Spin 30 Column, Tris, 100, includes 100 P-30 columns in Tris buffer, 100 buffer collection tubes, and instruction manual	
732-6226	Micro Bio-Spin 30 Column, Tris, 1,000, includes 1,000 P-30 columns in Tris buffer, 1,000 buffer collection tubes, and instruction manual	



732-6204	Empty Micro Bio-Spin Column, 100,
	includes 100 empty columns, 100 buffer col-
	lection tubes, and instruction manual



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