Aurum[™] Vacuum Manifold Instruction Manual



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Section 1 Introduction

The Aurum™ vacuum manifold and column adaptor plate (CAP)` comprise a system which allows a single vacuum manifold to be used with either 96-well plates or columns with luer ends. Separate manifolds are no longer required for different preparative formats; the Aurum vacuum manifold and CAP ensure fast, high-quality sample preparation in both formats, while maintaining the simplicity of handling associated with vacuum processing. The uniquely designed vacuum regulator allows for complete control of negative pressure within the Aurum vacuum manifold.

Section 2 Kit Components

The Aurum™ vacuum manifold contains the following components as shown in Figure 1:

- Vacuum manifold consisting of:
 - manifold top
 - manifold base
- Column adaptor plate
- Vacuum regulator
- Stage
- Luer caps
- Tubing (5/16" ID)

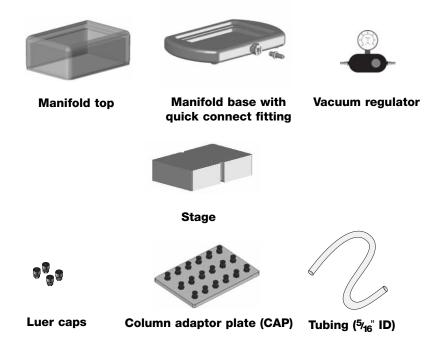


Fig. 1. Aurum vacuum manifold kit components.

Section 3 Care and Maintenance

The AurumTM vacuum manifold should be stored clean and dry at room temperature. After each use rinse with distilled water and wipe dry with paper towels or air dry. Never use alcohol or other organic reagents to clean the manifold. Never use abrasive materials to clean the manifold.

Gasket surfaces should be carefully rinsed to remove salts, which could inhibit contact between manifold parts and the column adaptor plate (CAP). Check the tubular and flat silicone gaskets for wear before and after use of the manifold. In the event that the bottom tubular gasket requires replacement, a length of $^{1}/_{8}$ " OD silicone tubing may be used.

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Section 4 Aurum™ Vacuum Manifold

The Aurum vacuum manifold is designed to accommodate the 96-well plates used in Bio-Rad's Aurum total RNA 96 kit. The manifold may also be compatible with other manufacturers' kits. For column processing, the column adaptor plate (CAP) is required.

The CAP allows the Aurum vacuum manifold to to be used for processing columns with a luer fitting. The CAP has 18 black luer fittings in a 6 x 3 array. Up to 18 Aurum miniprep columns can be accommodated on the CAP without the need for connectors or other manifold accessories. The CAP will also accommodate other columns with luer ends.

When vacuum is applied to the manifold, the CAP should self-seat, forming an airtight seal without the need to press down on the CAP. However, the application of gentle downward force may occasionally be required to facilitate seating.

The vacuum regulator is included and strongly recommended for measuring negative pressure, and for gradually increasing negative pressure during different protocols.

The Aurum manifold consists of two primary parts: the manifold base and top. The top has an attached tubular silicone gasket, allowing the manifold base and top to seal when a vacuum is introduced. Attached to the opening in the manifold top is a flat silicone gasket, enabling Aurum 96-well purification plates or the CAP to establish an airtight seal with the manifold top when a vacuum is applied. Collectively these two gaskets facilitate self-seating of all components, reducing the need to apply external pressure to the manifold to initiate vacuum. However, under certain conditions application of gentle downward force on the plates or CAP may be required to facilitate seating. In the event that the tubular silicone gasket becomes detached, it can easily be reinserted into the track on the underside of the manifold top.

Each Aurum vacuum manifold is supplied with a stage that centers and elevates a 96-well sample collection plate during elution from the purification plate. The stage should be placed with the printed "this side up" in the base with the groove side down to allow for airflow.

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Section 5 Vacuum Regulator

The vacuum regulator (see Figure 2) allows complete control of the negative pressures inside the Aurum™ vacuum manifold, allowing optimum levels of negative pressure for efficient sample preparation. This is particularly important when a gradual increase in negative pressure is required during 96-well plate purifications. Attached in-line between the vacuum source and filter flask, the regulator measures the difference between ambient pressure and pressure within the vacuum manifold in inches of mercury (inHg). The regulator is capable of measuring negative pressures up to −30 inHg. See Table 1 for pressure unit conversions.

At various points in the protocols, it is necessary to release the vacuum inside the manifold so that plates and columns can be removed. To release the negative pressure, rotate the regulator dial fully counter-clockwise until the gauge reads < 1 inHg. The vacuum is restored inside the manifold by rotating the control dial clockwise until the desired negative pressure is attained.

The regulator control dial is located off-center, toward the right side of the regulator. The vacuum source is attached to the right hose fitting and the vacuum manifold is attached to the left fitting (see Figure 3).

Table 1. Pressure unit conversions

To convert from inches of mercury (inHg) to:	Multiply by:
millimeters of mercury or torr (mmHg, torr)	25.4
millibar (mbar)	33.85
atmospheres (atm)	0.03342
pounds per square inch (psi)	0.4912
kilopascals (kPa)	3.385

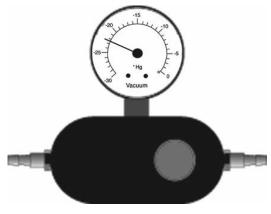


Fig. 2. Vacuum regulator and gauge.

Section 6 Vacuum Manifold Setup

Tubing provided is 4 ft long and must be cut in appropriate pieces before proceeding.

Prior to setup, you may ensure that the gauge pointer is adjusted to zero by removing the lens cover, followed by turning the adjustment pin located beneath the dial face.

Vacuum setup

- 1. Cut tubing into three pieces of appropriate length.
- 2. Use one piece of tubing to connect the vacuum source to the **right** side of the vacuum regulator.
- 3. Use another piece of tubing to connect the **left** side of the vacuum regulator to the sidearm of the filter flask.
- 4. Place a rubber stopper **with hole** into the mouth of the filter flask. Insert a serological pipette (or comparable) into the hole of the stopper.
- Snap in the black sealed end of the quick connect fitting into the manifold hase
- 6. Finally, use the last piece of tubing to connect the filter flask to the quick connect fitting of the nozzle of the manifold.

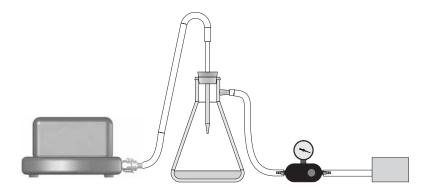


Fig. 3. Vacuum setup conditions.

Section 7 Use for 96-Well Plates

Manifold wash setup (see Figure 4A)

- Place the manifold top on the base, ensuring complete and uniform contact between the manifold top and base.
- 2. Place the desired 96-well purification plate on the manifold top and apply the recommended vacuum pressure for your application.

Manifold elution setup (see Figure 4B)

3. When eluting from the purification plate, place the stage on the manifold base, with the etched "this side up" facing up. Place a clean 96-well microtiter collection plate securely on top of the stage and replace the manifold top.

Note: If desired, waste can be collected into an optional pipet tip box lid instead of draining directly into the vacuum trap.

4. After use, rinse the Aurum™ vacuum manifold with water and air dry or wipe with paper towels. Failure to rinse the vacuum manifold at the end of each use will cause the manifold to become cloudy and pitted.

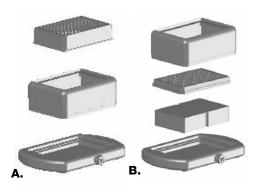


Fig. 4. Setup and use for 96-well plate processing.

A) 96-well wash; B) 96-well elution.

Section 8 Use for the Column Adaptor Plate (CAP)

Manifold wash setup (see Figure 5)

- 1. Insert the CAP (luer ends up) into the depression in the vacuum manifold top. Ensure that the CAP rests evenly on the gasket.
- 2. Insert the luer ends of the desired columns into the available luer fittings, ensuring a tight fit.
- Close the unused luer fittings with the caps provided. Close caps by rotating clockwise until light resistance is encountered. Excessive tightening of a cap may cause the luer fitting to dislodge when the cap is removed.
- 4. The manifold is now ready for column processing according to the vacuum protocol of the appropriate column purification kit.
- 5. When ready to elute, proceed with the appropriate spin elution step as recommended by the protocol.
- 6. After finishing the elution, rinse the CAP and Aurum vacuum manifold with water, and air dry or wipe with paper towels.

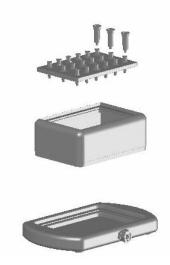


Fig. 5. Setup and use for column processing.

Section 9 Troubleshooting Guide

Problem	Possible Cause	Possible Solution
Insufficient negative pressure in vacuum manifold	Open vacuum regulator	Close vacuum regulator by turning completely clockwise
	Manifold top not seated properly on base	Ensure uniform contact between manifold top and base
	Purification plate or CAP not fully seated	Press down gently on the corners of the plate or CAP to seal
	Unused CAP luer fittings not closed completely	Close unused luer fittings completely with caps
	Gasket residue preventing even contact between manifold parts	Rinse manifold with distilled water
	Lower gasket misaligned	Press gasket back into groove
	Physical damage to manifold or CAP (e.g., chips and cracks)	Replace vacuum manifold or CAP
	Purge step in purification protocol	If closure of wells or columns (e.g., with sealing tape) restores negative pressure, no corrective action needed
Excessive noise during vacuuming	Purification plate or CAP not fully seated	Press down gently on plate or CAP to seat
	Gasket residue preventing even contact between manifold parts	Rinse manifold with distilled water
	Worn manifold gasket	Replace gasket

Problem	Possible Cause	Possible Solution
Incorrect negative pressure from vacuum regulator	Vacuum source connected to left side of vacuum	Connect vacuum to right side of vacuum
Clogging of purification plate or CAP columns	Insufficient negative pressure in vacuum manifold	See "Insufficient negative pressure in vacuum manifold" problem above

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Section 10 Ordering Information

Catalog #	Description
732-6470	Aurum Vacuum Manifold
732-6820	Aurum Total RNA Mini Kit
732-6830	Aurum Total RNA Fatty and Fibrous Tissue Kit
732-6800	Aurum Total RNA 96 Kit
732-6400	Aurum Plasmid Mini Kit