PELCO® Atomic Force Microscopy Gold Calibration Kit

Product. No. 16205

An important NEW product for AFM researchers, characterized colloidal gold particles for:

- Characterization of scanning tip geometry
- Reliable calibration of the vertical scale of piezoelectric response
- Characterizing vertical dimensions of co-absorbed biomolecules

Five sizes of colloidal gold particles are available in the kit. The kit contains 8 numbered 15mm AFM disks with mica attached for calibration and tip characterization. Any remaining colloidal gold can be used for co-absorption with biomolecules or other applications.

PELCO® AFM Gold Standard Kit Contains:

Product No.	
16214.....	PELCO® 15mm AFM Disk Carrier
16218-1.....	8 each numbered 15mm AFM Disks with 9.9mm Mica Disks
16220	PELCO® AFM Magnetic Probe
15700-AFM.....	500µl ~5nm Gold Colloid
15701-AFM.....	500µl ~15nm Gold Colloid
15702-AFM.....	500µl ~30nm Gold Colloid
15703-AFM.....	500µl ~10nm Gold Colloid
15704-AFM.....	500µl ~20nm Gold Colloid
18021.....	500µl 0.1% Poly-L-Lysine
16079-AFM.....	9 each Adhesive Tabs
Protocol and Reprint	

The five gold colloids contained in this kit have been characterized as follows:

See enclosed data sheets

PROTOCOL

1. Suggested dilutions of the gold colloids if used individually as a standard

   5nm and 10nm – Dilute 1:10 to 1:20 with double distilled deionized water (DI water) or other ultrapure water

   a. When used undiluted, a very high particle density will result

   b. There are approximately 1 x 10^{13} gold particles/ml (5nm) and 5.7 x 10^{12} gold particles (10nm) undiluted

   15nm and 20nm – Dilute 1:5 with DI water or other ultrapure water or use undiluted

   a. When used undiluted, a high particle density will result

   b. There are approximately 1.4 x 10^{12} gold particles (15nm) and 7 x 10^{11} gold particles/ml (10nm) undiluted

   30nm – Used undiluted – there are approximately 2 x 10^{11} gold particles undiluted

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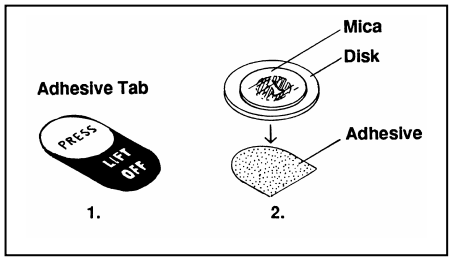
2. Suggested mix and dilution when using all three gold colloids on the same standard

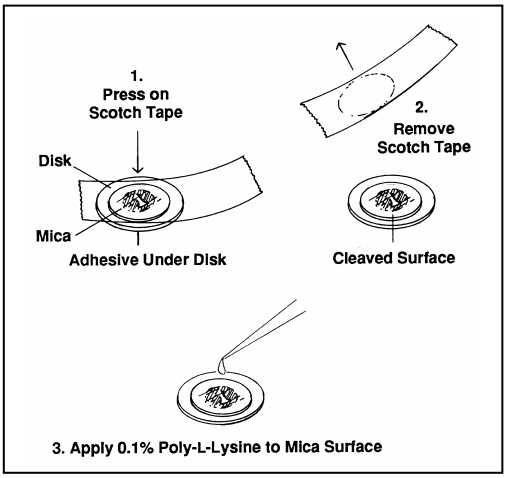
   a. Add 25µl of each size gold colloid to a 1.5ml microcentrifuge tube or similar tube
   b. Add 75µl of DI water and mix well
   c. Smaller quantities of the 5 and 15nm gold colloid can be used (i.e. 20µl) if desired

3. After the desired dilutions have been made, you are ready to begin making the actual standards.

   a. Place the 15mm disk, mica side up, on an adhesive surface. Nine adhesive tabs are supplied for this purpose. Simply place the **PRESS** portion of the tab on a countertop next to a sink and press down on the word "Press" and then pull the tab up. A thin layer of adhesive will be left on the countertop. Place the disk on the adhesive. This will hold the disk in place during the sample making process.



   b. With the disk in place on the adhesive, the mica can now be cleaved. It is easiest to use ½" wide Scotch Tape for this procedure, ¾" wide Scotch Tape will work, but not as well. Press the Scotch Tape (~3" long) to the surface of the mica disk and then smoothly remove the tape from the mica. The top layer of the mica will be removed by the adhesive on the tape. Continue to cleave the 9.9mm mica disk until the cleaved surface is mirror smooth to the eye (the center portion of the disk should be mirror smooth). Many times more than one cleave will be necessary. Good cleaves will produce an even spreading of the aqueous reagents in the forthcoming steps.



   c. After cleaving, apply 20µl of 0.1% Poly-L-Lysine to the mica surface. This should spread evenly across the whole mica surface. If it does not, it may be necessary to cleave the mica disk again. There are usually 20+ cleaves to a disk. Leave the Poly-L-Lysine on the surface for 30-60 seconds, then rinse with ~1ml of DI water and immediately dry the mica surface with nitrogen, argon or some other dry gas (20-40 lbs. line pressure works well).

   d. After drying immediately apply 20µl of the gold colloid solution to the mica surface and let incubate for five minutes. Like the Poly-L-Lysine, the gold should spread evenly across the cleaved mica surface. NOTE: The colloid can be left on longer than 5 minutes. If this is done, the particle density will increase greatly.

e. Rinse with ~1ml DI water and immediately dry with nitrogen, argon or some other dry gas. The easiest and safest way to remove the 15mm disk from the adhesive is to use a single edge razor blade to cut the disk off the adhesive. Be careful not to flip the disk over during the removal process. The disk can be placed upside down in the disk carrier at this point.

f. Incubate the disk (in the carrier) in a 60°C oven for one hour to overnight prior to scanning with the AFM. After removal from the oven, the standard should be stored in a desiccator.

IMPORTANT NOTES:

1. The standards should be scanned under conditions of low humidity (relative humidity <20%).

2. Probe forces of ~15nN work well with blunt or dull tips (radius of curvature ~25-75nm). With sharp tips (radius of curvatures <25nm) the probe force should be kept small (<10nN) or the gold colloids may be swept off the surface.

3. Not all attempts at making a standard will be successful; therefore, 8 disks with mica have been supplied. It has been our experience that 50% of the time a usable standard will result from this protocol.