

## Instructions for Thiol PEG Reagents

### I. Introduction

The BroadPharm Thiol PEG is a thiol-terminated polyethylene glycol (PEG)-containing reagent with either a methyl ether, hydroxyl or carboxylic acid group at the other end. These reagents have defined molecular weights and spacer lengths and are used for modifying surfaces such quantum dots, self-assembled monolayers and magnetic particles. Functionalization of solid surfaces with PEG spacers significantly reduces nonspecific protein binding.

### II. Product Information

Storage: Upon receipt store desiccated at -20°C.

- The Thiol PEG reagents are low-melting solids or liquid that are difficult to weigh and dispense. To facilitate handling, make a stock solution by dissolving the reagent in dimethylsulfoxide (DMSO) or dimethylformamide (DMF).
- Store unused stock solution at -20°C. Equilibrate reagent vial to room temperature before opening to avoid moisture condensation. To minimize air exposure, keep the stock solution under an inert gas such as argon or nitrogen. Cap the stock solution with a septum and use a syringe to remove the solution.
- If the Carboxyl-Thiol PEG reagent is used for surface binding and further protein loading, the reagent-to-surface ratio in the reaction affect the number of carboxylic acid residues available for further modification. Optimize these ratios to obtain the modification level needed for the specific application.
- Use non-amine-containing buffers at pH 7-9 such as PBS (20mM sodium phosphate, 150mM NaCl; pH 7.4); 100mM carbonate/bicarbonate; or 50mM borate. Do not use buffers that contain primary amines, such as Tris or glycine, which compete with acylation.

### III. Procedures for Nanoparticle surface antibody conjugation with Thiol PEG

1. Equilibrate the Thiol PEG reagent to room temperature before opening bottle.
2. Prepare stock solutions by dissolving 100mg of each reagent in the desired amount of DMF or DMSO.
3. Prepare the appropriate amount of nanoparticle surface and desired amount of the Thiol PEG reagent in Phosphate-buffered saline (PBS, 20mM sodium phosphate, 0.15M NaCl, pH 7.2).
4. Incubate the reaction for 2 hours at room temperature and then wash the surface with PBS buffer to remove excess reagent.

*\*\*If using the Carboxyl-Thiol PEG reagent, please proceed to step 5-8.*

5. For Carboxyl-Thiol PEG reagent: the newly introduced carboxylic acid groups can be activated by adding appropriate amounts of EDC and NHS to the modified surface in MES-buffered saline (0.1M MES, 0.5M NaCl; pH 6.0 or 0.1M MES, 0.9% NaCl; pH 4.7) and reacting for 15 minutes at room temperature.
6. Wash the surface with MES-buffered saline to remove any remaining EDC and NHS.
7. Add the desired amine-containing substrate (prepared in PBS buffer) to the activated surface and react for 2 hours at room temperature.
8. Add hydroxylamine or another amine-containing buffer to quench the conjugation reaction.