

# Carboxylic acid-SAM Formation Reagent

## General Information

Carboxylic acid-SAM Formation Reagent is applied for the preparation of carboxylic acid terminated self-assembled monolayers (SAMs) on a gold surface of biosensors such as for QCM (Quartz Crystal Microbalance), SPR (Surface Plasmon Resonance) or electrical analysis. Carboxylic acid-SAM can be used as an interface to immobilize a protein or a peptide on a gold surface coupled with carboxyl group activation method using NHS/WSC. Amine Coupling Kit (Product code: A515) is available for protein immobilization on the gold surface coated with a carboxylic acid-SAM.

Carboxylic acid-SAM Formation Reagent is especially designed to minimize a non-specific protein adsorption on the surface.

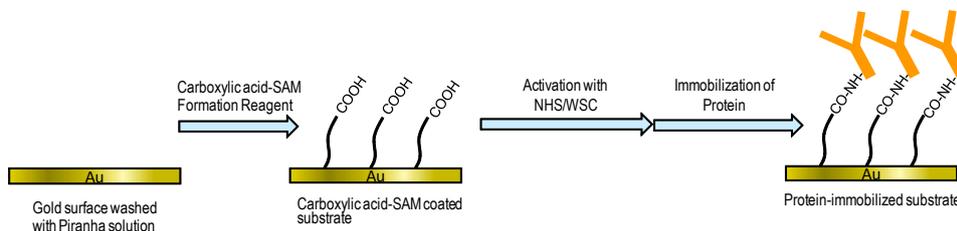


Fig.1 Preparation of a biosensor using Carboxylic acid-SAM Formation Reagent

## Contents

1  $\mu\text{mol} \times 3$

## Caution

Since a trace amount of colorless or slightly yellowish liquid is in the tube, please centrifuge prior to use.

## Storage

0-5 °C

## General Protocol for Carboxylic acid-SAM Preparation

Preparation of a Carboxylic acid-SAM surface on a gold substrate

- 1) Add 1 ml ethanol to a tube, and pipette to prepare 1 mmol/l Carboxylic acid-SAM solution. <sup>a)</sup> Then, dilute the solution 10-fold with ethanol for Step 2).
- 2) Immerse a gold substrate <sup>b)</sup> in the reagent solution prepared at Step 1) at room temperature and leave it overnight.
- 3) Wash the substrate several times with ethanol and purified water sequentially. <sup>c)</sup>

a) If the reagent hardly dissolve by pipetting, use an ultrasonic bath. Use freshly prepared reagent solution at Step 2).

b) Clean the gold substrate with Piranha solution, if necessary. Mix 3 volumes of sulfuric acid and one volume of hydrogen peroxide solution to prepare Piranha solution. Since the solution is severely caustic, handle with great care.

c) Store the SAM-coated substrate under nitrogen gas in a tightly sealable glass container at 0-5 °C.

## General Protocol for Protein Immobilization

This product does not include necessary reagents and solutions for a protein immobilization. Amine Coupling Kit (Product code: A515) is available for the protein immobilization on the carboxylic acid-SAM surface. If not use the kit, follow this general protocol.

- Required reagents and buffers-

- N-Hydroxysuccinimide (NHS)
- 1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide (WSC)
- pH4-6 buffer solution
  - \* MES buffer is generally used.
- pH8-9 buffer solution
  - \* Carbonate buffers, phosphate buffers or good's buffers are generally used.

- 1) Prepare 100 mmol/l NHS and 100 mmol/l WSC solutions using pH4-6 buffer solution .
- 2) Mix equal volume of NHS and WSC solutions immediately, and add to the carboxylic acid-SAM coated gold substrate.
- 3) Leave it at room temperature for 1 hr, and wash several times with pH4-6 buffer solution.
- 4) Add a protein solution <sup>a)</sup> to the substrate, and leave it at room temperature for 1 hr.
- 5) Wash with PBS, and go to blocking process with a blocking solution <sup>b)</sup> if necessary.

a) The solution should be prepared with a buffer solution (pH8-9). Use 10-1,000  $\mu\text{g}/\text{ml}$  of protein solution for immobilization.

b) One mol/l ethanolamine solution (pH8.5) is commonly used for blocking.