SGNP (Sugar chain immobilized Gold Nano Particle)

SGNPs

Cat. No.	PRODUCT NAME	Cat. No.	PRODUCT NAME
G-00M-100	Alpha-Glucosyl-GNP	G-00C-100	Beta-Glucosyl-GNP
G-00E-100	Alpha-Galactosyl-GNP	G-00L-100	Beta- Galactosyl -GNP
G-AGN-100	Alpha-GlcNAc-GNP	G-BGN-100	Beta-GlcNAc-GNP
G-AAN-100	Alpha-GalNAc-GNP	G-BAN-100	Beta-GalNAc-GNP
G-0AF-100	Alpha-Fucosyl-GNP	G-0BF-100	Beta-Fucosyl-GNP
G-AMA-100	Alpha-Mannosyl-GNP		
G-03S-100	3SialylGalactosyl -GNP	G-06S-100	6SialylGalactosyl -GNP

*One vial contains each different SGNP showing Abs_{530nm} = 3.0[/cm] when the whole SGNP is dissolved in 1 mL of buffer.

SGNP set

Cat. No.	PRODUCT NAME	Content
G-AB1-250	SGNPs #1	Contains following 11 kinds of SGNPs in a set. Alpha-Glucosyl-GNP; Beta-Glucosyl-GNP; Alpha-Galactosyl-GNP; Beta-Galactosyl–GNP; Alpha-GlcNAc-GNP; Beta-GlcNAc-GNP; Alpha-GalNAc-GNP; Beta-GalNAc-GNP; Alpha-Fucosyl-GNP, Beta-Fucosyl-GNP, Alpha-Mannosyl-GNP
G-AB2-250	SGNPs #2	Contains following 13 kinds of SGNPs in a set. Alpha-Glucosyl-GNP; Beta-Glucosyl-GNP; Alpha-Galactosyl-GNP; Beta-Galactosyl–GNP, Alpha-GlcNAc-GNP; Beta-GlcNAc-GNP; Alpha-GalNAc-GNP, Beta-GalNAc-GNP, Alpha-Fucosyl-GNP; Beta-Fucosyl-GNP; Alpha-Mannosyl-GNP; 3SialylGalactosyl –GNP; 6SialylGalactosyl –GNP

*Each vial in a set contains SGNP showing Abs_{530nm} = 3.0[/cm], when the whole SGNP is dissolved in 0.25 mL of buffer

Properties

SGNP（Sugar chain - immobilized Gold Nano-Particle） is a gold nano particles immobilized with structurally defined sugar chains, showing red-purple color (λmax = ca. 530 nm). SGNP is a very convenient tool for evaluating sugar chain – protein interaction, since the interaction can be detected visually, and can be quantified from the change of OD at 530 nm. There are several applications as follows:

1. Aggregation of protein
2. Inhibition assay
3. Dot-blotting
4. Isolation and purification of lectin from a crude extract

*SGNP is a reagent for research. Do not use for other purposes.

Delivery as

Lyophilized powder

Stability

Lyophilized powder can be stored within a year at room temperature.

Amount of SGNP in a vial

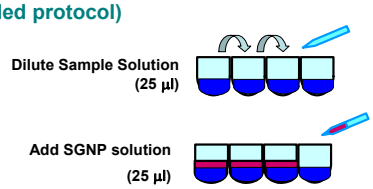
Cat. No. G-*-100**
Abs_{530nm} = 3.0[/cm] (dissolved in 1 mL of buffer)
Cat. No. G-*-250**
Abs_{530nm} = 3.0[/cm] (dissolved in 0.25 mL of buffer)

Addition of a reducing agent, such as mercaptoethanol, may affect the properties of SGNP.

Application#1: Aggregation assay of protein(s)

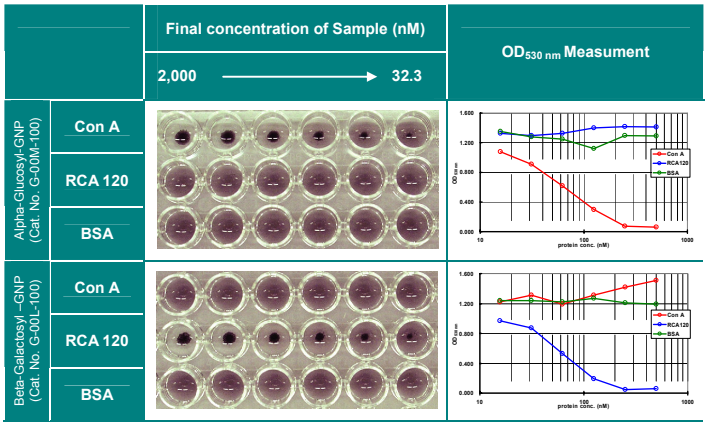
Regularly sugar chain binding proteins, such as lectins, possess multiple binding sites for sugar chains. By the addition of SGNP to the protein solution, the protein may form aggregates with sugar chains immobilized SGNP in a short time of period. The change can be seen visually, or can be quantified by measuring OD at around 530 nm. Using SGNP, the binding properties (selectivity, dissociation constant (K_D), specificity, etc.) are easily evaluated.

(Recommended protocol)



1. Dissolved SGNP at Abs_{530nm} ＝ 3.0 using your buffer.
   *see Amount of SGNP
2. In wells of 96-well microtiter plate (round bottom), 25µl of protein solution was added with changing the concentration.
   *Sequential 1:2 dilution from ca. 200 µg/ml (or ca. 4 µM), 6 points or more.
3. Addition of 25 µl of SGNP solution prepared as above. Gentle agitation for 0.5 to 2 hr at room temperature.
4. Measure OD at 530 nm of the supernatant.

(Example)

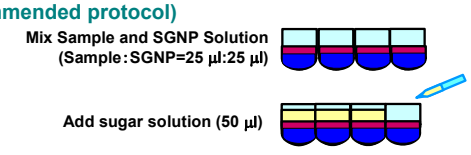


* Con A<Concanavalin A> (EY LABORATORIES, Cat. No.L1104250)
RCA120<Ricinus communis Agglutinin I > (Vector, Cat. No.L-1080)
BSA <Albumin, bovine serum> (SIGMA Aldrich, Cat. No.A0281)

Application#2: Inhibition assay

This assay is useful to know the specificity of the ligands for the target protein. By the addition of inhibitor (mono-saccharide, oligo-saccharide, mimetic compounds, glycoprotein, or drug candidate), the formed protein-SGNP aggregates may be re-dissolved by the competitive binding of protein with the inhibitor.

(Recommended protocol)



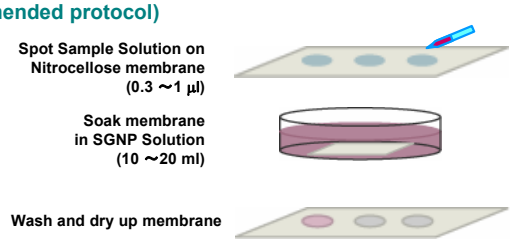
1. Dissolve SGNP at Abs_{530nm} ＝ 4.0 ～ 6.0 using your buffer
   *see Amount of SGNP
2. In wells of 96-well microtiter plate (round bottom), 25μl of protein solution (Conc. > K_D) and 25 μl of SGNP solution as prepared above were added.
   *It is important to know K_D value of the protein against sugar chain on SGNP
3. 50 μl of the inhibitor dissolved in the same buffer (conc. 0.1 ～ 50 mM) is added to the above mixture. Then, agitate for 1 hr or overnight.
4. Measure OD at 530 nm of the supernatant.

Application#3: Probe for Dot-Blotting assay

The sugar chain binding potency of the trace amount of your samples can be detected.

* Independent on the valency of sugar-binding proteins

(Recommended protocol)



1. Prepare 10～20ml SGNP solution of Abs_{530nm} ＝ 0.15 ～ 0.30 using your buffer.
   *see Amount of SGNP
2. On nitrocellulose membrane* (10 x 30 mm), spot your sample (0.3 – 1 μL, 0.1- 2.0 μg) and dry up at room temperature.
   *Trans-Blot™ Transfer Medium, Pure Nitrocellose Membrane (0.2 μm) (Bio-Rad, Cat. No.162-0146)
3. To 10 mL of SGNP solution in a glassware (φ 50 mm), the membrane is soaked with a gentle agitation for 5 – 30 min.
   *You may need to check the staining every 5 min. Please anchor the membrane with tweezers
4. Wash the membrane with buffer and dry up.
   *If you see too high background, decrease the concentration of SGNP or shorten the soaking time.

(Example)

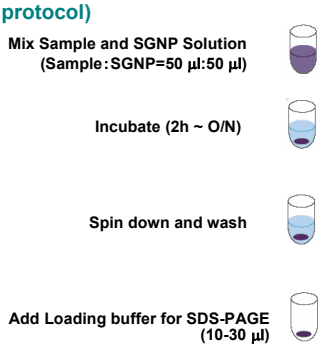
SGNP \ Sample	Con A	WGA	RCA 120
Alpha-Glucosyl-GNP (Cat. No. G-00M-100)			
Beta-Galactosyl-GNP (Cat. No. G-00L-100)			
Beta-GlcNAc-GNP (Cat. No. G-AGN-100)			

* Con A<Concanavalin A> (EY LABORATORIES, Cat. No.L1104250)
RCA120 <Ricinus communis Agglutinin I > (Vector, Cat. No.L-1080)
WGA (Wheat Germ) (Seikagaku Kogyo, Cat. No.5400)

Application#4: Isolation and identification of target protein from a crude extract

Quick isolation and identification of protein(s) having multi-binding sites (lectin) from a plant-derived crude extract of cell-lysate are available using SGNP. Once you get an aggregate by mixing appropriate SGNP with your extract or lysate, the aggregate can be directly applied for SDS-PAGE. Then, the protein band(s) in SDS-gel is analyzed according to the proteomics procedure.

(Recommended protocol)



1. Dissolve SGNP at Abs_{530nm} ＝ 3.0 using your buffer
   *see Amount of SGNP
2. To 50 μl of SGNP solution in a 1.5 mL of eppendorf tube, add 50 μl of your extract. The mixture is incubated for 1 h to overnight at 4 degree C with a gentle agitation.
   * The protein concentration in the extract may be in the range between 500 and 10 mg/mL.
3. Centrifugation at 6,000 ～ 10,000 x g for 10 min, and remove the supernatant.
4. To the precipitate, add 500 μl of buffer, vortex for 10 sec and centrifuge at 6,000 ～ 10,000 x g for 10 min, and remove the supernatant. This may repeat 2 more times.
5. To the precipitate, add 10-30 μl of sample preparation buffer*, boiled up for 10 min..
   *Laemmli Sample Buffer (Bio-Rad, Cat. No.161-0737)
6. SDS-PAGE
   *Reducing or non-reducing condition can be used.

(Example)