

FAQ – Cline Scientific

Ordering and Support

How to order

Contact your distributor, or Cline Scientific directly if your country is not represented. Use our web shop or order form on www.clinescientific.com

Distributors

Sweden (headquarters) and the Nordic countries

Cline Scientific AB
Carl Skottsbergs g. 22B
413 19 Gothenburg
Sweden
Tel: +46(0)313875555
E-mail: order@clinescientific.com

California: Iwai North America Inc

USA East Coast: Mancel Associates Inc

Japan: Funakoshi Co., Ltd

Germany: BioTrend GmbH

Czech Republic: Pentagen s.r.o.

For contact information to distributors, please visit the Cline Scientific home page.

Technical Support

For technical support contact Cline Scientific directly.

We will answer your questions as soon as possible, usually within 24 hours.

Tel: +46(0)313875555

E-mail: info@clinescientific.com

Opening Hours: 8-16 GMT+1

Production

The production is located in Sweden.

Delivery

The Nano Gradient and Nano Surfaces are delivered in cold packs and should be transferred to fridge conditions as soon as possible according to separate handling guidelines.

The Cline Nano Particles are delivered in standard packages and should be stored cold.

Products

Nano Particles

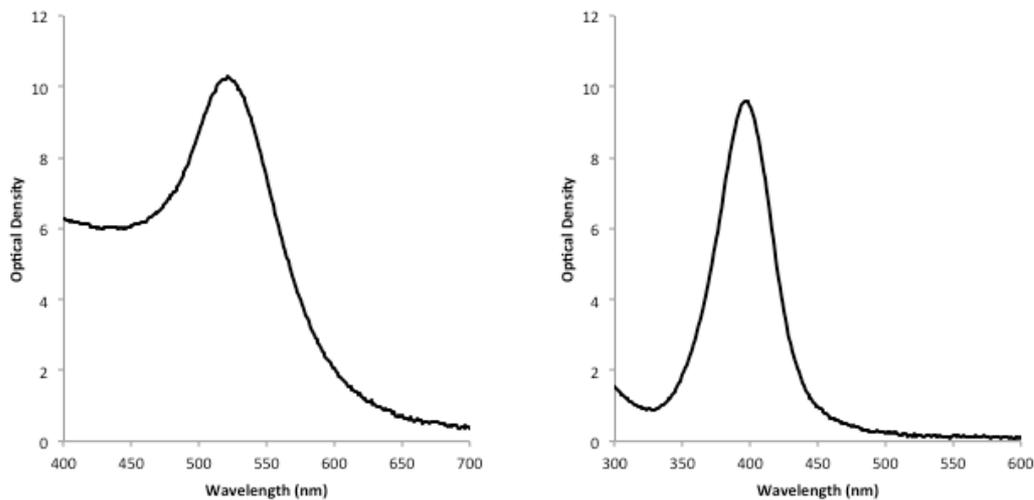
Quality control

In what way does Cline Scientific test the quality of the produced batches?

The products are subjected to ocular inspection, UV-VIS spectrometry, and electron microscopy (TEM).

How is the concentration of particles in solution determined?

The concentration is calculated from the amount of gold or silver ions that are used in the production reaction, which reaches completion. The UV-VIS spectrum is documented showing height (correlating to particle concentration) and shape (correlating to particle size).



Typical UV-VIS spectra for AuNP 20nm (left) and AgNP 20nm (right) (for more information; see Product Sheets)

Shelf Life

How should the particles be stored?

In the fridge, 4-8 °C

How long is the shelf life of particles in 4 °C?

When stored in 4 °C the UV-VIS spectrum of our gold nanoparticles (AuNP) will not change in a year, and for our silver nanoparticles (AgNP) 2 years.

In the 5nm AuNP (NP01-0051010) a brownish precipitate may appear after some months of storage. The precipitate is possible to dissolve upon shaking and the spectrum does not change. If black flakes are found the product should be discarded. The latter has not been observed within the recommended shelf life.

What are the Storage Procedures?

AgNP can be stored short periods at room temperature and in ambient light; however, storage at lower temperatures prolongs the shelf life and prolonged exposure to light can change the material size or shape.

AuNP products are recommended to be stored in 4 °C.

PEG-ylated AuNP and AgNP should be stored in 4 °C in *glass bottles* for optimal stability.

Is the particles light sensitive?

AuNP is not light sensitive; however AgNP may be more sensitive to certain wavelengths of light.

In which solutions are the particles stable?

MilliQ-water is the best choice. 10nm AuNP will aggregate in salt solutions of more than 10nM.

Cline's PEG-coated AuNP and AgNP will be stable in salt solutions of at least 1M and in cell culture media with and without serum.

Can the particles be stored in the freezer?

If frozen, the nanoparticles will irreversibly aggregate and the solution color will change.

PEG-coated particles can be frozen without aggregation; however we recommend storing them in 4 °C.

Usage

What are the Handling Procedures?

Shake the bottle prior to use. During storage the nanoparticles may settle to the bottom. Prior to using the nanoparticles, re-suspend the settled nanoparticles by vigorously shaking the bottle until a homogenous solution is obtained. Inspect the bottom of the bottle to ensure that no settled precipitates remain.

If precipitates remain, see: "*Can I use the product after its expiration date?*"

Can I use the product after its expiration date?

If using the product after expiration date or if storage did not follow our guidelines, a small amount of metal plating may develop on the sides or bottom of bottles. If plating is observed, place the bottle in a bath sonicator for no more than 30 seconds. Remove and shake the bottle. If plating is still visible, wait 30 seconds before sonicating again for <30 seconds (to limit excessive heating of the particles). Repeat as necessary until plating is no longer visible. If sonication is needed, please perform the quality control.

Quality Control:

Look for any dark-colored particulates or flakes floating in the solution, a decrease in the solution's color intensity, or a shift in the solution's color. If any of these are observed, the materials should be analyzed via, e.g., UV-Vis spectroscopy or TEM for quality verification.

How is PEG bound to the particles?

With a covalent thiol bond

How large are the particles with PEG-coating?

PEG-5000 adds approx. 10nm to the radius of the nanoparticle core.

Is autoclavation possible?

No

Services

Can you functionalize or modify gold- and silver nanoparticles with my molecule?

Yes. We can offer this service. Send a request to your distributor or Cline Scientific directly with your specifications.

Nano Gradient and Nano Surfaces

Quality control

In what way does Cline Scientific test the quality of the produced batches?

The products are subjected to ocular inspection, and electron microscopy (SEM).

How can it be verified that biomolecules are bound to the Nano Gradients and Nano Surfaces?

Some examples of techniques to use are mass spectroscopy (TOF-SIMS) and imaging surface plasmon resonance (iSPR). For more information see scientific publications on the Cline Scientific web page.

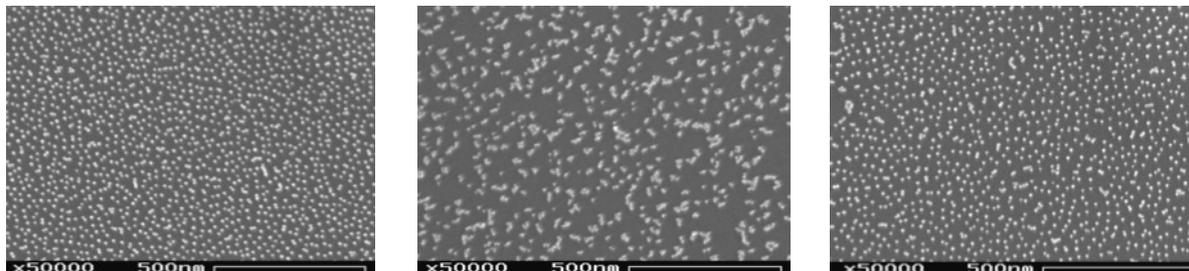
Shelf Life

How long can the surfaces be stored?

Un-functionalized Nano Gradients and Nano Surfaces (NG06-0109999 and NS06-010109999) will last for at least 6 months at 4 °C. No change in distribution of nanoparticles on the surfaces can be observed during this timeframe.

What happens at storage in different conditions?

Gradient surfaces (NG06-0109999) were stored in different conditions for one month. In 1) water at 4 °C, 2) in water in the freezer, and 3) dried out. As visible in the images below, it is recommended that the surfaces are stored in water or PBS at 4 °C where no change in nanoparticle distribution is observed. Other storage may alter the surface on the nano-level, mainly causing nanoparticle aggregation.



One month storage in 1) 4 °C water, 2) freezer, and 3) dried out

How long will a functionalized surface last in storage?

The time is very much dependent on the molecule/biomolecule itself and for how long time the molecule attached can last in different conditions. The concentration of attached molecule is very low since they are present on the surface in monolayers. What the exact shelf life will be differs from molecule to molecule, but in general it will be in range of several weeks.

Usage

How long will a functionalized surface last in cell culture?

The surfaces have been documented to work perfectly in cell culture conditions for at least two weeks.

How long have surfaces been stored before usage?

It has been reported from customers that functionalized Nano Gradient Surfaces have been stored in fridge conditions between 1 day and 1 week before usage.

Are there potentially cell toxic substances on the cell culture surfaces?

No, the chemistry used to attach AuNP and biomolecules have not shown to affect cell viability

Which fixations can be used on the surface when fixing cells after experiments?

PFA (4-5%) has successfully been used, incubated for 24 hours. It is important to take care not to rinse the cells off the surface during the fixation and subsequent rinsing process.

Can I stain the cells grown on the Cline Nano Gradient and Cline Nano Surface?

Antibodies, immunofluorescence and standard staining techniques works well

Can I use a microscope to analyze my cell results?

Yes, the surfaces work with most microscopes. For further information, see product descriptions.

Will the image quality be affected by the purple color from nanoparticles?

In microscopes the purple color will not affect the image quality or color. In fact, the Nano Gradient and Nano Surface have a glass substrate with a superior image quality compared to cell culture plastics.

Will my cells attach to the surface? I have had problems with glass surfaces before

The nano technology allows biomolecules (such as ECM proteins e.g. laminins) to bind very well to the glass and will make cells attach to the surface, unlike the case with untreated glass surfaces where even coating procedures can be difficult.

How many cells should be seeded on the surfaces?

It is important to not seed too many cells. Too many cells may result in bad image resolution when studying cell responses to the surfaces and may also result in cells releasing from the surface during fixing and rinsing.

Fibroblasts were incubated for 24h on the surfaces. Approximately 10-50 000 cells per cm² was successfully seeded.

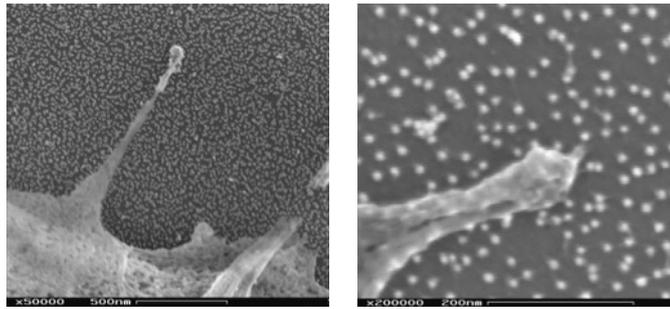
In experiments with Induced Pluripotent Stem cells (IPS cells), 30-100 000 cells per cm² was tried successfully.

Will the proteins stay if you wash with e.g. buffer?

Yes, however, use caution and don't touch the surface.

Will the particles stay on the surface during cell experiments?

Yes, as an example; After growing fibroblast cells on the Cline Nano Gradient surface and studying the result using SEM, there were no evidence of cells causing AuNPs to aggregate, change position, or in any other way altering the surface structure.

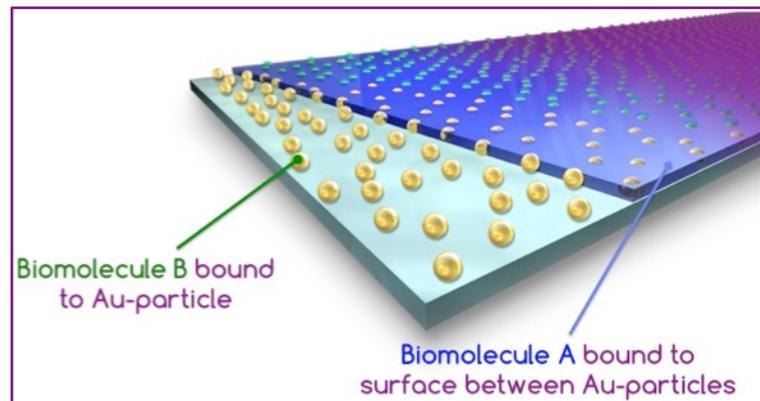


SEM images showing fibroblasts growing on the Cline Nano Gradient surface. The cell-binding RGD-peptide was bound to AuNP and PEG covered the glass surface between the particles.

How is it possible to modify the Cline Nano Gradient or Nano Surface with two kinds of biomolecules such as e.g. proteins and growthfactors.

This can be done by binding, e.g., growth factors to the AuNP (B), allowing for larger proteins, such as laminins (A), to be attached to the space between particles.

Cline Scientific can provide this service for customers, or the customers can do it themselves using biotinylation and a protocol provided by Cline. For further information, see user guides on the home page.



How do different sizes and shapes of biomolecules affect the surface gradient?

This depends very much on the biomolecules used and the method to bind them to the surfaces, however; in general terms the following can be said:

Small molecules (<1000 kDa) may be approximately 10 on each AuNP. Larger molecules, such as large peptides and whole proteins, can be ca 1-4 per particle depending on, e.g., its shape. A big fibrous protein can cover several particles, thus decreasing the gradient effect but creating gradual protein stiffness instead. Long proteins, such as laminin, collagen fibers etc., are bound to the glass surface between the particles for the best gradient effect.

Services

Can you functionalize or modify the Cline Nano Gradient or Nano Surface with my own molecules?

Yes. We can offer this service. Send a request to your distributor or Cline Scientific directly with your specifications.