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NANOGOLD® and FluoroNanogold™ Conjugates

Quick Facts

Storage upon receipt:
- 2–6°C
- Do not freeze
- Protect from light

Abs/Em: See Table 1

Notes: Avoid exposure to high temperatures (>37°C) and to thiols (e.g., β-mercaptoethanol or dithiothreitol)

Introduction

In collaboration with Nanoprobes, Inc., Molecular Probes offers both unlabeled NANOGOLD® conjugates and Alexa Fluor dye–labeled FluoroNanogold™ conjugates (Table 1). NANOGOLD conjugates consist of streptavidin or Fab′ fragments of affinity-purified goat anti–mouse IgG, goat anti–rabbit or rabbit anti–goat IgG antibodies covalently conjugated to uniform, 1.4 nm–diameter gold clusters. The gold particles are present in a ~1:1 ratio with the streptavidin or Fab′ fragment. In Fab′ fragments, the NANOGOLD particle is attached in the hinge region, remote from the antigen-binding site. Our FluoroNanogold conjugates are NANOGOLD conjugates (described above) labeled with an Alexa Fluor dye. NANOGOLD and FluoroNanogold conjugates can be used as probes in immunoblotting, light microscopy or electron microscopy. FluoroNanogold conjugates can also be used with fluorescence microscopy.

NANOGOLD conjugates represent a new technology that has distinct advantages over the use of traditional colloidal-gold probes. NANOGOLD conjugates are smaller and offer better penetration than other commercially available gold probes.[1] Also, whereas colloidal gold is negatively charged and can bind nonspecifically to proteins, resulting in high background signals or false-positive results, NANOGOLD conjugates exhibit low nonspecific binding and minimal background signals. NANOGOLD conjugates are stable at a wide range of pHs and ionic strengths and have a shelf life of up to one year. Researchers prefer NANOGOLD conjugates over colloidal gold for high-resolution immunoelectron microscopy, for labeling nucleic acids and other non-protein species, or where the target antigen is in a hard-to-reach region of the cell. Furthermore, NANOGOLD conjugates develop better with silver than do most colloidal golds, giving them higher sensitivity. Silver enhancement can be used to make the immunolabeling useful for electron microscopy, light microscopy and immunoblotting, with improved results.

FluoroNanogold conjugates offer all of the superior characteristics of the NANOGOLD conjugates, including excellent penetration and stability, as well as the properties conferred by our premium Alexa Fluor dyes, which include superior brightness and photostability. Unlike colloidal golds, which can quench fluorescence, the presence of the NANOGOLD cluster has a negligible effect on fluorescence emission. Research has shown that FluoroNanogold conjugates provide unprecedented correlation between fluorescence and electron microscopy data.[2]

Materials

Contents

The NANOGOLD and FluoroNanogold reagents are supplied in unit sizes of 1 mL as 80 µg protein/mL solutions in phosphate-buffered saline (PBS), pH 7.4, containing 0.1 % BSA and 0.05% sodium azide.

Storage

When these products are stored undiluted at 2–6°C, they are stable for at least three months. DO NOT FREEZE. Store NANOGOLD and FluoroNanogold reagents protected from light.

Table 1. *NANOGOLD and FluoroNanogold conjugates.*

Conjugate	Abs/Em *	Fab′ fragment of goat anti–mouse IgG	Fab′ fragment of goat anti–rabbit IgG	Fab′ fragment of rabbit anti–goat IgG	Streptavidin
NANOGOLD	NA/NA	N-24915	N-24916	N-24917	N-24918
Alexa Fluor488 FluoroNanogold	495/519	A-24920	A-24922	A-24924	A-24926
Alexa Fluor 594 FluoroNanogold	590/617	A-24921	A-24922	A-24925	A-24927

* Absorption (Abs) and approximate fluorescence emission (Em) maxima, in nm. NA = Not applicable.

THIOL CAUTION: NANOGOLD and FluoroNanogold particles degrade upon exposure to concentrated thiols such as β-mercaptoethanol or dithiothreitol. If such reagents must be used, concentrations should be kept below 1 mM and exposure restricted to 10 minutes or less.

TEMPERATURE CAUTION: NANOGOLD and FluoroNanogold particles are generally quite stable;[3] however, under some conditions NANOGOLD particle– or FluoroNanogold particle–labeled specimens or conjugates may not be stable above 50°C. Best results are obtained at room temperature or 2–6°C, and 37°C incubations should be avoided. Use low-temperature embedding media (e.g., Lowicryl) if labeling before embedding,[4] and do not bake tissue blocks with NANOGOLD or FluoroNanogold particles. If your experiment requires higher-temperature embedding, then silver enhancement should be performed before embedding.

Application

NANOGOLD and FluoroNanogold probes have far better penetration into permeabilized cells and nuclei than do colloidal-gold probes. NANOGOLD Fab′ conjugates and FluoroNanogold Fab′ conjugates are even smaller than immunofluorescent IgG labels. The NANOGOLD streptavidin conjugate is especially useful for nucleic acid detection. There is absolutely no aggregation with NANOGOLD probes, and they are very stable, because the NANOGOLD particle is covalently linked to the biomolecule. With a subsequent brief (5–15 min) silver enhancement (LI Silver Enhancement Kit, L-24919), NANOGOLD particles give a signal visible even in ordinary brightfield optical microscopes (UV optics are not necessary), with a sensitivity that is usually better than fluorescence.

Since the same NANOGOLD probe can be used for the EM as well, preliminary staining can be followed in the light or confocal microscope while a parallel sample can be examined at the ultrastructural EM level. This flexibility is not available with fluorescent probes. With FluoroNanogold probes, the preliminary observations can be made by fluorescence microscopy.

Extensive application notes are available at the Nanoprobes Web site: www.nanoprobes.com.

References

1. J Histochem Cytochem 43, 329 (1995); **2.** J Histochem Cytochem 45, 631 (1997); **3.** *Immunogold-Silver Staining: Principles, Methods and Applications, M. A. Hayat, Ed., CRC Press (1995) pp. 71–96.* **4.** *Immunogold-Silver Staining: Principles, Methods and Applications, M. A. Hayat, Ed., CRC Press (1995) pp. 57–69.*

Product List

Current prices may be obtained from our Web site or from our Customer Service Department.

Cat #	Product Name	Unit Size
A-24920	Alexa Fluor® 488 FluoroNanogold™ Fab′ fragment of goat anti-mouse IgG *80 µg protein/mL*	1 mL
A-24922	Alexa Fluor® 488 FluoroNanogold™ Fab′ fragment of goat anti-rabbit IgG *80 µg protein/mL*	1 mL
A-24924	Alexa Fluor® 488 FluoroNanogold™ Fab′ fragment of rabbit anti-goat IgG *80 µg protein/mL*	1 mL
A-24926	Alexa Fluor® 488 FluoroNanogold™ streptavidin *80 µg protein/mL*	1 mL
A-24921	Alexa Fluor® 594 FluoroNanogold™ Fab′ fragment of goat anti-mouse IgG *80 µg protein/mL*	1 mL
A-24923	Alexa Fluor® 594 FluoroNanogold™ Fab′ fragment of goat anti-rabbit IgG *80 µg protein/mL*	1 mL
A-24925	Alexa Fluor® 594 FluoroNanogold™ Fab′ fragment of rabbit anti-goat IgG *80 µg protein/mL*	1 mL
A-24927	Alexa Fluor® 594 FluoroNanogold™ streptavidin *80 µg protein/mL*	1 mL
L-24919	LI Silver (LIS) Enhancement Kit	1 kit
N-24915	NANOGOLD® Fab′ fragment of goat anti-mouse IgG *80 µg protein/mL*	1 mL
N-24916	NANOGOLD® Fab′ fragment of goat anti-rabbit IgG *80 µg protein/mL*	1 mL
N-24917	NANOGOLD® Fab′ fragment of rabbit anti-goat IgG *80 µg protein/mL*	1 mL
N-24918	NANOGOLD® streptavidin (streptavidin, NANOGOLD® conjugate) *80 µg protein/mL*	1 mL

Contact Information

Further information on Molecular Probes' products, including product bibliographies, is available from your local distributor or directly from Molecular Probes. Customers in Europe, Africa and the Middle East should contact our office in Leiden, the Netherlands. All others should contact our Technical Assistance Department in Eugene, Oregon.

Please visit our Web site — **www.probes.com** — for the most up-to-date information

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NANOGOLD and FluoroNanogold conjugates of streptavidin and Fab' fragments are prepared for Molecular Probes by Nanoprobes, Inc.