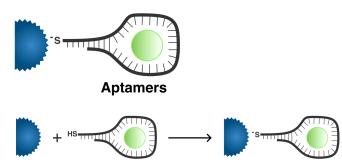


## PRODUCT DATA SHEET

### AptamerREADY™ Gold NanoUrchin Conjugation Kit



#### Description

Cytodiagnostics AptamerREADY<sup>™</sup> Gold NanoUrchin conjugation kits have been optimized for high efficiency onestep conjugation of thiolated aptamers directly to the gold NanoUrchin surface.

The kit contains ready-to-use pre-made mixtures. No activation, manipulation, or time consuming "salt-aging" steps are required for conjugation. Simply mix your reduced thiol-modified aptamer with the supplied pre-activated gold NanoUrchins. Conjugation of the aptamer is achieved by the formation of a strong and stable gold-thiol bond without any additional linkers.

#### **Features & Benefits**

- Allows conjugation of aptamers to gold NanoUrchins with sizes between 50nm-100nm.
- Fast and convenient one-step conjugation reaction with no pre-activation requirements or manipulation of the gold NanoUrchins.
- No time-consuming "salt-aging" procedures.
- Results in a thiol-aptamer conjugated directly to the gold NanoUrchin surface without any linkers.
- Optimized for use in crosslinking based lateral flow applications.

#### **Gold NanoUrchin Specifications**

**Gold surface:** Proprietary AptamerREADY<sup>™</sup> coating **Core diameter:** Available with diameters from 50nm-100nm **Optical density (OD):** OD=2 when the contents of each vial are dissolved to a final volume of 1 ml.

Particles per ml: Core size dependant, please see table II. Lambda max: Core size dependant, please see table II.

#### Storage

Store at -20° C. Stable for at least 3 months if stored as specified.

#### **Product Safety and Handling**

This product is for R&D use only, not for drug, household, or other uses. Please review the material safety datasheet (MSDS) available online for proper safety and handling procedures.

#### Procedure

## Reduction of thiol-modified aptamers (e.g. trityl-S-S-Aptamer)

1. Prepare a 0.15 M sodium phosphate buffer, pH 8.5 supplemented with 0.1 M DTT.

**Note:** pH is important for proper reduction of oligonucleotide.

- 2. Dissolve lyophilized aptamer to a final concentration of 500  $\mu M$  in H2O.
- 3. Mix 50 μl of dissolved aptamer with 450 μl sodium phosphate buffer.
- 4. Incubate 1-2 hours at room temperature to reduce aptamer.
- Separate reduced aptamer from trityl-SH and DTT using a NAP 5 column operated in H<sub>2</sub>O, GE Healthcare.
- 6. Final eluate from NAP 5 column will be 1ml in H<sub>2</sub>O with an approximate concentration of 25  $\mu$ M.

**Note:** Exact concentration of final eluate can be measured with UV-VIS spectroscopy by measuring the absorbance at 260nm.

# Conjugation of thiolated aptamer to AptamerREADY™ gold NanoUrchins

- 1. Resuspend one vial of lyophilized AptamerREADY™ gold NanoUrchins with 740 μl of H<sub>2</sub>O.
- 2. Transfer into a 1.5 ml microcentrifuge tube.

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 Add 160 μl of reduced thiolated aptamer at 1 μM (0.0075 nmol/μl)\* in H<sub>2</sub>O as prepared above and incubate for at least 1 hour at room temperature.

**Note:** 1 µM aptamer is a good starting concentration, but if aggregation or poor sensitivity is observed, the following Aptamer concentrations can be attempted for a given NanoUrchin size range (based on a 30nt oligonucleotide):

| Particle size (nm) | 50-70   | 80-100 |  |  |
|--------------------|---------|--------|--|--|
| [aptamer] (µM)     | 0.25-10 | 0.1-5  |  |  |

- 4. Add 100 μl of 1M NaCl.
- Incubate for at least 1 hour at room temperature to allow binding of the aptamer to the gold NanoUrchin surface.

**Note:** Longer incubation times may improve surface coverage.

- Centrifuge at the appropriate speed for your particular gold NanoUrchin size (see table I) for 30 minutes to pellet your aptamer gold NanoUrchin conjugate.
- 7. Remove supernatant.
- Resuspend conjugate in 200 μl of storage buffer. The optical density of the particles should be 10 if a 100% recovery has been achieved.

Common storage buffer: 10 mM sodium phosphate buffer, pH 7.0, 100 mM NaCl and 0.01% (w/v) NaN<sub>3</sub>.

- 9. Measure optical density with a spectrophotometer and adjust concentration as desired.
- 10. Store conjugate at +4°C.



*Figure 1.* 0.5% (*w*/*v*) agarose gel analysis of 60nm AptamerREADY<sup>™</sup> gold NanoUrchins before and after functionalization with a thiolated aptamer (30 bases). Gel was operated at 100V in 0.5X TBE buffer for 30 minutes.

**Table I.** Appropriate G forces for centrifugation of gold NanoUrchins. Note that recommended conditions are for a volume of 1ml and centrifugation using a microcentrifuge.

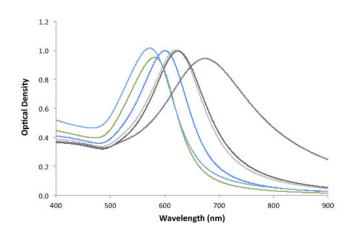
| Size (nm) | Speed (g) | Time (min) |  |
|-----------|-----------|------------|--|
| 50        | 2,000     | 30         |  |
| 60        | 1,125     | 30         |  |
| 70        | 700       | 30         |  |
| 80        | 600       | 30         |  |
| 90        | 500       | 30         |  |
| 100       | 400       | 30         |  |

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**Table II.** Gold NanoUrchin specifications by size. Please note that all values below are indicated at an optical density of 1 ( $OD/cm^{-1}$ ) at their respective lambda max. At other optical densities the values need to be adjusted (e.g. NPS/ml (@OD2) = 2 x NPS/ml (@OD2)).

| Diameter<br>(nm) | Peak SPR<br>Wavelength<br>(nm) | NPS/mI   | Wt. Conc.<br>(mg/ml) | Molar Ext<br>(M <sup>-1</sup> cm <sup>-1</sup> ) | Size<br>Dispersity<br>(+/-nm) | Particle<br>Volume<br>(nm <sup>3</sup> ) | Surface<br>Area<br>(nm²) | Surface/<br>Volume<br>Ratio | Particle<br>Mass (g) | Molar Mass<br>(g/mol) | Molar Conc. |
|------------------|--------------------------------|----------|----------------------|--|-------------------------------|--|--------------------------|-----------------------------|----------------------|-----------------------|-------------|
| 50               | 560                            | 3.51E+10 | 4.45E-02             | 1.72E+10   | <8%                           | 6.54E+04                                 | 7.85E+03                 | 0.12                        | 1.27E-15             | 7.64E+08              | 5.83E-11    |
| 60               | 585                            | 1.96E+10 | 4.30E-02             | 3.07E+10   | <10%                          | 1.13E+05                                 | 1.13E+04                 | 0.1                         | 2.19E-15             | 1.32E+09              | 3.25E-11    |
| 70               | 600                            | 1.20E+10 | 4.17E-02             | 5.03E+10   | <10%                          | 1.80E+05                                 | 1.54E+04                 | 0.086                       | 3.48E-15             | 2.10E+09              | 1.99E-11    |
| 80               | 620                            | 7.82E+09 | 4.06E-02             | 7.70E+10   | <10%                          | 2.68E+05                                 | 2.01E+04                 | 0.075                       | 5.20E-15             | 3.13E+09              | 1.30E-11    |
| 90               | 630                            | 5.37E+09 | 3.97E-02             | 1.12E+11   | <8%                           | 3.82E+05                                 | 2.54E+04                 | 0.067                       | 7.40E-15             | 4.46E+09              | 8.92E-12    |
| 100              | 670                            | 3.84E+09 | 3.89E-02             | 1.57E+11   | <8%                           | 5.24E+05                                 | 3.14E+04                 | 0.06                        | 1.02E-14             | 6.11E+09              | 6.37E-12    |
|                  |                                |          |                      |  |                               |  |                          |                             |                      |                       |             |



| Catalogue Number | Description  | Sizes                      |
|------------------|--|----------------------------|
| AUC-50-X*        | 50nm AptamerREADY Gold NanoUrchin Conjugation Kit  | 3 reactions & 10 reactions |
| AUC-60-X*        | 60nm AptamerREADY Gold NanoUrchin Conjugation Kit  | 3 reactions & 10 reactions |
| AUC-70-X*        | 70nm AptamerREADY Gold NanoUrchin Conjugation Kit  | 3 reactions & 10 reactions |
| AUC-80-X*        | 80nm AptamerREADY Gold NanoUrchin Conjugation Kit  | 3 reactions & 10 reactions |
| AUC-90-X*        | 90nm AptamerREADY Gold NanoUrchin Conjugation Kit  | 3 reactions & 10 reactions |
| AUC-100-X*       | 100nm AptamerREADY Gold NanoUrchin Conjugation Kit | 3 reactions & 10 reactions |

\*X Indicates quantity, i.e. -1 for a 3 reaction kit and -2 for a 10 reaction kit

For custom sizes and information on bulk quantities and prices please contact our customer service department.

#### **Ordering Information**

For ordering call 866-344-3954 or visit us online.