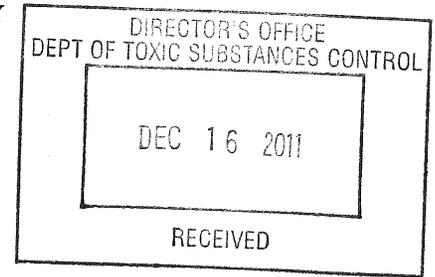




STANFORD UNIVERSITY

ENVIRONMENTAL HEALTH & SAFETY



December 14, 2011

Maziar Movassaghi, Acting Director
Department of Toxic Substances Control
1001 "I" Street
P.O. Box 806
Sacramento, California 95812-9806

Re: DTSC Information Call In: Chemical Information and Analytical Test Methods for
Nanometals, Nanometal Oxides, and Quantum Dots

Dear Mr. Movassaghi,

Stanford University respectfully submits the following information regarding the use of specified Nanometals, Nanometal Oxides, and Quantum Dots in research at Stanford University as requested on January 4, 2011 from the Department of Toxic Substances Control as required by California Health and Safety Code sections 57018-57020.

1. Summary of Business and Certification Information

Stanford University is an institution of higher education and research. Attachment 1 provides the requested Business and Certification information for Sections A-C.

2. Summary of Nanomaterial Chemical & Physical Properties; Analytical Test Methods

Overview:

Thirteen academic research laboratories at Stanford University have been identified as using the following nanomaterials of interest in eighteen research projects throughout the campus: Nano Silver, Nano Titanium Dioxide, Nano Zinc Oxide, and Quantum Dots. Attachments 2-5 provide the information requested in Sections D-F for each of the four nanochemicals.

The referenced research laboratories and projects are within the departments of Chemical Engineering, Chemistry, Material Sciences & Engineering, Geology, Applied Physics, Mechanical Engineering, and Cellular & Molecular Physiology. They were identified using data generated from three sources; a prior 2009 campus-wide nanomaterials research use survey, an inventory search of Stanford's electronic chemical materials inventory system (ChemTracker), and a search of Stanford websites. Principal Investigators were asked to complete Sections D-F, and site visits of selected laboratories and phone interviews by EH&S staff were used to gather additional information, as necessary.

Nano Zero Valent Iron and Nano Cerium Oxide usage at Stanford University have not been reported.

Description of Use:

Stanford University uses the above nanomaterials in the basic and applied research areas of biomedical applications involving microscopy imaging, energy storage devices, fuel production, and fundamental materials research. References and website information for selected published and copyrighted research projects are included Attachments 2-5. These sources provide additional information on the nature of the nanomaterials research involving the referenced materials taking place at Stanford University. (Note that due to publisher copyright protection concerns, entire articles are not provided in this document, but the location for access to such reference information is provided.)

Researchers at Stanford University use nanomaterials on the laboratory-scale¹. Those academic research laboratories that synthesize nanomaterials range in total amount produced from 0.2 milligrams to 25 grams annually. Labs reported that they purchase between 10 milligrams to 150 grams total annually; the majority of quantum dots are purchased as kits and contain nominal quantities of nanomaterials. Material Safety Data Sheets are provided for purchased materials.

Analytical Methods:

a. For Chemical and Physical Properties:

Depending on the nature of the research, the laboratory may rely on information provided by the vendor or conduct quantitative or qualitative assessments to determine the specific physical or chemical parameter. On campus, the analytical methods to evaluate the various physical and chemical properties of each of the four types of nanomaterials indicated in Attachments 2-5 include:

- Transmission Electron Microscopy (TEM)
- Dynamic Light Scattering (DLS)
- X-ray photoelectron spectroscopy (XPS)
- Near-infrared (NIR)
- Scanning Electron Microscopy (SEM)
- Brunauer-Emmett-Teller (BET) Surface Area Analysis
- Inductively coupled plasma optical emission spectrometry (ICP-OES)
- Energy-dispersive X-ray spectroscopy (EDX)

Reseachers perform these standard laboratory techniques primarily at Stanford's core analytical facilities including:

- Stanford Nanocharacterization Laboratory
<http://www.stanford.edu/group/snl/>
- Stanford University Soft and Hybrid Facilities Materials Laboratory
<http://www.stanford.edu/group/snc/equipment/soft%20materials.html>
- Small Animal Imaging Facility
<http://mips.stanford.edu/aboutus/facilities/clark/sci3/index.html>
- Stanford Nanofabrication Facility
<http://snf.stanford.edu>

The references for selected published research projects included Attachments 2-5 provide additional regarding the analytical methods employed (see the Experimental Methods section in

¹ Per Cal/OSHA 8 CCR 5191, Occupational Exposures to Hazardous Chemicals in Laboratories, laboratory scale refers to the work with substances in which the containers used for reactions, transfers, and other handling of substances are designed to be easily and safely manipulated by one person. "Laboratory scale" excludes workplaces whose function is to produce commercial quantities of materials.

the respective articles). Product information from Invitrogen on their Quantum Dot products are also included, which include experimental protocols for the use of these commercial kits.

b. For Environmental Matrices:

Six research projects involve the evaluation of Quantum Dots, Nano Zinc Oxide, and Nano Silver in water and cells using various methods of analysis including fluorescence microscopy, spectrophotometry, and electron microscopy. Other environmental matrices of DTSC interest are reportedly not evaluated in current research activities (i.e., air, soil, sludge, chemical waste, fish, blood, adipose tissue, urine).

Comment on Future Rule Making

If rulemaking regarding the use, fate, and transport of the specified Nanometals, Nanometal Oxides, and Quantum Dots is pursued in the future, Stanford University would support the continued partnership between the DTSC and California institutions of higher education to provide continued input. Our goal is to ensure that any future rulemaking uses a risk-based approach that considers the small laboratory scale utilized in the course of education and academic research at universities and colleges throughout California.

Kindly direct any inquiries regarding this response to Lawrence M. Gibbs, Associate Vice Provost for Environmental Health and Safety, lgibbs@stanford.edu, (650) 723-7403.

Sincerely,



Lawrence M. Gibbs, CIH
Associate Vice Provost

Attachments:

- Attachment 1: Section A-C, Chemical, Business Identification Information and Certification
- Attachment 2: Section D-F for Quantum Dots (w/MSDSs, references for selected publications, and Invitrogen Experimental Protocols)
- Attachment 3: Section D-F for Nano Silver (w/MSDS and references for selected publications)
- Attachment 4: Section D-F for Nano Titanium Dioxide (w/MSDSs and references for selected publications)
- Attachment 5: Section D-F for Nano Zinc Oxide Dots

Attachment 1:

**Section A-C, Chemical, Business Identification Information and
Certification**



STANFORD UNIVERSITY

ENVIRONMENTAL HEALTH & SAFETY

Section A: Chemical(s) (check each one which applies to your company)

- | | | |
|---|---|--|
| <input checked="" type="checkbox"/> Nano Silver | <input checked="" type="checkbox"/> Nano Titanium Dioxide | <input type="checkbox"/> Nano Cerium Oxide |
| <input type="checkbox"/> Nano Zero Valent Iron | <input checked="" type="checkbox"/> Nano Zinc Oxide | <input checked="" type="checkbox"/> Quantum dots |

Section B: Business Identification Information (check one and complete items 1 - 10)

- | | | | | |
|--|--|--|--|--|
| <input type="checkbox"/> Sole Owner | <input type="checkbox"/> Corporation | <input type="checkbox"/> Limited Liability Company (LLC) | <input type="checkbox"/> Limited Liability Partnership (LLP) | <input type="checkbox"/> Unincorporated Business Trust |
| <input type="checkbox"/> Spouse's Co-Ownership | <input type="checkbox"/> Registered Domestic Partnership | <input type="checkbox"/> General Partnership | <input type="checkbox"/> Limited Partnership | <input checked="" type="checkbox"/> Other: (describe) University |

1. Name of Sole Owner, Corporation, Partnership, Institution, Other.

Board of Trustees of the Leland Stanford Junior University

2. Business Trade Name ("Doing Business As," if any)

N/A

3. Business Address (physical location of your business: street number and name, city, state, country, zip or postal code)

480 Oak Rd., Stanford, CA, USA 94305-8007

4. Mailing Address (street name and number, P.O. box, city, state, country, zip or postal code, if different from 3)

Same as above

5. Business Website Address(es):

www.stanford.edu; ehs.stanford.edu

6. Name of Owner, Responsible Corporate Officer, Partner, Other

Lawrence Gibbs for John Hennessy, President

7. Contact Information for Person 6 above:

Name:

Lawrence Gibbs

Business Telephone:

(650) 723-7403

Title:

Associate Vice Provost for EH&S

Email:

lgibbs@stanford.edu

8. Number of Employees (California employees).

10,233

9. NAICS Code(s) for this business:

Primary:

923110

Other:

Other:

10. Nano Chemical Business Type: (check applicable)



Manufacturer



Importer



Researcher

Section C: Certification (for this complete submittal)

I am duly authorized to prepare and submit this information, as a formal response to the request pursuant to Health and Safety Code section 57019(d)(1), and certify the information and statements made herein, and in the attachments, are correct to the best of my knowledge and belief.

Name:

Lawrence Gibbs,
Associate Vice Provost for
Environmental Health and
Safety
Stanford University

Signature:

Date:

12/14/2011

Attachment 2:

Section D-F for Quantum Dots

Section D: Nanomaterial Chemical and Physical Properties

Product/ Productions Information (based on eight Stanford University academic research laboratories)			
Nano Chemical Name:		Quantum Dots	
Commercial Name(s):		Purchased materials- <ul style="list-style-type: none"> • Qdot 605 Streptavidin Conjugate • Qdot 655 Streptavidin Conjugate • Qdot 705 Streptavidin Conjugate • Qdot incubation buffer • Qdot 705 ITK carboxyl quantum dots • QTracker 655 Cell Labeling Kit • Qdot 605 ITK Streptavidin Conjugate • Qdot 625 Streptavidin Conjugate • PbS Core EviDots Note: Two non-commercial quantum dot materials in use (AgS, MoS2)	
Annual Production Volume:		On-campus synthesized volume: 25g Materials externally produced/ supplied: <ul style="list-style-type: none"> • ~10 kits Invitrogen QDot Kits • 15 ml Pbs EviDots • 15 milligrams AgS 	
Production Method(s):		On-campus synthesis: Solution phase self-assembly (for MoS2)	
Identification of the Supplier(s):		<ul style="list-style-type: none"> • Invitrogen • Evident Technologies • Suzhou Institute of Nanotech and Nanobionics, Chinese Academy of Sciences 	
Physical Properties			
Parameter	Value / Range ¹ (include units)		Name of Analytical Method(s) ²
Shape (Morphology)	Sphere, ellipsoid		<ul style="list-style-type: none"> • Scanning Electron Microscopy • Transmission Electron Microscopy
Density	0.002- 7.23 g/cm ³		None (Theoretical, calculated based on mass balance)
Surface Area	MoS2: 50-1000 m ² /g		<ul style="list-style-type: none"> • Brunauer, Emmet and Teller Method • Scanning Electron Microscopy • Transmission Electron Microscopy
	Others: Unknown		Unknown
Particle Size Distribution	Air	Unknown	Unknown
	Liquid	2-20 nm	<ul style="list-style-type: none"> • Dynamic Light Scattering • Scanning Electron Microscopy • Transmission Electron Microscopy
	Solid/ Powder	AgS: 10.2±0.4 nm	Transmission Electron Microscope
		MoS2: 1 um-mm clusters	Visible inspection
		Others: Unknown	Unknown
Other (specify)			
Chemical Properties			

Parameter		Value / Range ¹ (include units)	Name of Analytical Method(s) ²
Chemical Composition		<ul style="list-style-type: none"> • CdSe/ZnS (Qdots) • PbS (EviDot) 	Unknown (from supplier)
		<ul style="list-style-type: none"> • AgS • MoS₂ 	X-Ray Photoelectron Spectroscopy
Surface Modification (Coating, Functionalization)		Carboxylate/ streptavidin (Qdots)	Unknown (from Supplier)
		Mercaptopropionic acid (for AgS)	Oil-water dual phase extraction and exchange
		Oleic acid, poly(styrene)-poly(vinylpyridine) (for MoS ₂)	Result of synthesis
Purity		MoS ₂ : > 99%	X-Ray Photoelectron Spectroscopy
		Others: Unknown	Unknown
Surface Charge		MoS ₂ : 0	None (theoretical)
		Others: Unknown	Unknown
Dispersion:	Air	MoS ₂ : Agglomerates into um-mm size clusters	Visible Observation; Transmission Electron Microscopy
		Others: Unknown	Unknown
	Liquid	MoS ₂ : Well dispersed in organic solvents	Visible Observation; Transmission Electron Microscopy
		Others: Unknown	Unknown
	Solid	MoS ₂ : Agglomerates into um-mm size clusters	Visible Observation; Transmission Electron Microscopy
		AgS: Agglomerated into chunks of ~1 mm	Visible Observation
Others: Unknown		Unknown	
Identifying and Determining Concentration of Nano Chemical, Its Metabolites, and Degradation Products in Specified Matrices Water, Air, Soil, Sediment, Sludge, Chemical Waste, Fish, Blood, Adipose Tissue, Urine, Other (Specify)		See Section F	
Solubility	Water Solubility	MoS ₂ : Poorly soluble	UV-Vis Transmission of optical density
		Others: Unknown	Unknown
	Solubility in Organic Solvent	MoS ₂ : Highly soluble	UV-Vis Transmission of optical density
		Others: Unknown	Unknown
n-Octanol- Water Partition Coefficient		Unknown	Unknown

Stability and Reactivity	Flammability	MoS ₂ : Solvent may be flammable	Based on solvent used in dispersion (e.g., Toluene)
		Others: NFPA Fire 0-3 (rating based on solvent)	None (from Supplier)
	Explosiveness	MoS ₂ : Solvent may be explosive	Based on solvent used in dispersion (e.g., Toluene)
		Others: Not indicated	Unknown
	Oxidizing Properties	MoS ₂ : Does not oxidize, but can be oxidized	Based on solvent used in dispersion (e.g., Toluene)
		Others: Unknown	Unknown
	Oxidation Reduction Potential	AgS: -0.88V for Ag ₂ S/Ag pair	Reference information
		MoS ₂ : +0.6V vs. reversible hydrogen electrode	Electrochemical cyclic voltammetry
		Others: Unknown	Unknown
	Storage Stability and Reactivity (Container Material)	Qdots: Stable, dark, plastic bottle (for product performance)	None (From Supplier)
		EviDot: Unknown	None (From Supplier)
		AgS, MoS ₂ : Stable, glass or plastic vial	Visible Observation
	Stability to Thermal, Sunlight, and Metal(s)	Qdots: Avoid thermal and sunlight exposure	N/A
		EviDot: Unknown	N/A
		AgS: Quantum yield decreases when exposed to sunlight	Near infrared fluorescence measurement
		MoS ₂ : Stable	Visible Observation

Section E: Globally Harmonized System Safety Data Sheet (SDS) or Materials Safety Data Sheet (MSDS)

<i>Chemical Name</i>	<i>Source</i>
Qdot 605 Streptavidin Conjugate Qdot 655 Streptavidin Conjugate Qdot 705 Streptavidin Conjugate Qdot incubation buffer Qdot 705 ITK carboxyl quantum dots QTracker 655 Cell Labeling Kit Qdot 605 ITK Streptavidin Conjugate Qdot 625 Streptavidin Conjugate	Invitrogen
PbS Core Evi Dots	Evident Technologies

Section F: Describe the analytical test method(s) that you use or plan to use to sample, prepare and analyze a specific matrix (water, air, soil, sediment, sludge, chemical waste, fish, blood, adipose tissue, and urine) to determine the identity and concentration of each specified nanomaterial.

<i>Nanomaterial</i>	<i>Purpose (sample, prepare, analyze)</i>	<i>Matrix</i>
Quantum Dot Nanocrystals	Analyze	Water, cell/ tissue cultures
Method description		
Three labs use Quantum dots used in cell/ tissue cultures. One lab is applying the material in water. Researchers are using various methods of analysis including but not limited to fluorescence microscopy, spectrophotometry, and electron microscopy.		

MATERIAL SAFETY DATA SHEETS

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code Q10101MP
Product name Qdot® 605 streptavidin conjugate

Company/Undertaking Identification

INVITROGEN CORPORATON
5791 VAN ALLEN WAY
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
5250 MAINWAY DRIVE
BURLINGTON, ONT
CANADA L7L 6A4
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS**Hazardous/Non-hazardous Components**

The product contains no substances which at their given concentration, are considered to be hazardous to health. We recommend handling all chemicals with caution.

3. HAZARDS IDENTIFICATION**Emergency Overview**

The product contains no substances which at their given concentration, are considered to be hazardous to health

Form
Liquid

**Principle Routes of Exposure/
Potential Health effects**

Eyes	No information available
Skin	No information available

3. HAZARDS IDENTIFICATION

Inhalation No information available
Ingestion No information available

Specific effects

Carcinogenic effects No information available
Mutagenic effects No information available
Reproductive toxicity No information available
Sensitization No information available

Target Organ Effects

No information available

HMIS

Health	0
Flammability	0
Reactivity	0

4. FIRST AID MEASURES

Skin contact Wash off immediately with plenty of water
Eye contact Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion Never give anything by mouth to an unconscious person
Inhalation Move to fresh air
Notes to physician Treat symptomatically.

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media Dry chemical
Special protective equipment for firefighters Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions Use personal protective equipment
Methods for cleaning up Soak up with inert absorbent material.

7. HANDLING AND STORAGE

Handling No special handling advice required
Storage Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection In case of insufficient ventilation wear suitable respiratory equipment
Hand protection Protective gloves
Eye protection Safety glasses with side-shields
Skin and body protection Lightweight protective clothing.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313

This product is not regulated by SARA.

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)

This product does not contain HAPs.

U.S. State Regulations

California Proposition 65

This product does not contain chemicals listed under Proposition 65

WHMIS hazard class:

Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

This material is sold for research and development purposes only. It is not for any human or animal therapeutic or clinical diagnostic use. It is not intended for food, drug, household, agricultural, or cosmetic use. An individual technically qualified to handle potentially hazardous chemicals must supervise the use of this material.

The above information was acquired by diligent search and/or investigation and the recommendations are based on prudent application of professional judgment. The information shall not be taken as being all inclusive and is to be used only as a guide. All materials and mixtures may be present unknown hazards and should be used with caution. Since Invitrogen Corporation cannot control the actual methods, volumes, or conditions of use, the Company shall not be held liable for any damages or losses resulting from the handling or from contact with the product as described herein. THE INFORMATION IN THIS MSDS DOES NOT CONSTITUTE A WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR ANY PARTICULAR PURPOSE.

End of Safety Data Sheet

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code Q10061MP B
Product name Qdot® incubation buffer

Company/Undertaking Identification

INVITROGEN CORPORATON
5791 VAN ALLEN WAY
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
5250 MAINWAY DRIVE
BURLINGTON, ONT
CANADA L7L 6A4
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS**Hazardous/Non-hazardous Components**

The product contains no substances which at their given concentration, are considered to be hazardous to health. We recommend handling all chemicals with caution.

3. HAZARDS IDENTIFICATION**Emergency Overview**

The product contains no substances which at their given concentration, are considered to be hazardous to health

Form
Liquid

**Principle Routes of Exposure/
Potential Health effects**

Eyes
Skin

No information available
No information available

3. HAZARDS IDENTIFICATION

Inhalation No information available
Ingestion No information available

Specific effects

Carcinogenic effects No information available
Mutagenic effects No information available
Reproductive toxicity No information available
Sensitization No information available

Target Organ Effects No information available

HMIS

Health	0
Flammability	0
Reactivity	0

4. FIRST AID MEASURES

Skin contact Wash off immediately with plenty of water
Eye contact Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion Never give anything by mouth to an unconscious person
Inhalation Move to fresh air
Notes to physician Treat symptomatically.

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media Dry chemical
Special protective equipment for firefighters Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions Use personal protective equipment
Methods for cleaning up Soak up with inert absorbent material.

7. HANDLING AND STORAGE

Handling No special handling advice required
Storage Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection In case of insufficient ventilation wear suitable respiratory equipment
Hand protection Protective gloves
Eye protection Safety glasses with side-shields
Skin and body protection Lightweight protective clothing.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313

This product is not regulated by SARA.

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)

This product does not contain HAPs.

U.S. State Regulations

California Proposition 65

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WHMIS hazard class:

Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

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The above information was acquired by diligent search and/or investigation and the recommendations are based on prudent application of professional judgment. The information shall not be taken as being all inclusive and is to be used only as a guide. All materials and mixtures may be present unknown hazards and should be used with caution. Since Invitrogen Corporation cannot control the actual methods, volumes, or conditions of use, the Company shall not be held liable for any damages or losses resulting from the handling or from contact with the product as described herein. THE INFORMATION IN THIS MSDS DOES NOT CONSTITUTE A WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR ANY PARTICULAR PURPOSE.

End of Safety Data Sheet

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code Q21361MP
Product name Qdot® 705 ITK™ carboxyl quantum dots

Company/Undertaking Identification

INVITROGEN CORPORATON
5791 VAN ALLEN WAY
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
5250 MAINWAY DRIVE
BURLINGTON, ONT
CANADA L7L 6A4
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

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3. HAZARDS IDENTIFICATION**Emergency Overview**

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Form
Liquid

**Principle Routes of Exposure/
Potential Health effects**

Eyes	No information available
Skin	No information available

3. HAZARDS IDENTIFICATION

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Ingestion No information available

Specific effects

Carcinogenic effects No information available
Mutagenic effects No information available
Reproductive toxicity No information available
Sensitization No information available

Target Organ Effects No information available

HMIS

Health	0
Flammability	0
Reactivity	0

4. FIRST AID MEASURES

Skin contact Wash off immediately with plenty of water
Eye contact Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion Never give anything by mouth to an unconscious person
Inhalation Move to fresh air
Notes to physician Treat symptomatically.

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media Dry chemical
Special protective equipment for firefighters Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions Use personal protective equipment
Methods for cleaning up Soak up with inert absorbent material.

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Handling No special handling advice required
Storage Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection In case of insufficient ventilation wear suitable respiratory equipment
Hand protection Protective gloves
Eye protection Safety glasses with side-shields
Skin and body protection Lightweight protective clothing.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

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Hazard Class	No information available
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The above information was acquired by diligent search and/or investigation and the recommendations are based on prudent application of professional judgment. The information shall not be taken as being all inclusive and is to be used only as a guide. All materials and mixtures may be present unknown hazards and should be used with caution. Since Invitrogen Corporation cannot control the actual methods, volumes, or conditions of use, the Company shall not be held liable for any damages or losses resulting from the handling or from contact with the product as described herein. THE INFORMATION IN THIS MSDS DOES NOT CONSTITUTE A WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR ANY PARTICULAR PURPOSE.

End of Safety Data Sheet

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code Q10121MP
Product name Qdot® 655 streptavidin conjugate

Company/Undertaking Identification

INVITROGEN CORPORATON
5791 VAN ALLEN WAY
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
5250 MAINWAY DRIVE
BURLINGTON, ONT
CANADA L7L 6A4
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS**Hazardous/Non-hazardous Components**

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3. HAZARDS IDENTIFICATION**Emergency Overview**

The product contains no substances which at their given concentration, are considered to be hazardous to health

Form
Liquid

Principle Routes of Exposure/**Potential Health effects**

Eyes
Skin

No information available
No information available

3. HAZARDS IDENTIFICATION

Inhalation No information available
Ingestion No information available

Specific effects

Carcinogenic effects No information available
Mutagenic effects No information available
Reproductive toxicity No information available
Sensitization No information available

Target Organ Effects No information available

HMIS

Health	0
Flammability	0
Reactivity	0

4. FIRST AID MEASURES

Skin contact Wash off immediately with plenty of water
Eye contact Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion Never give anything by mouth to an unconscious person
Inhalation Move to fresh air
Notes to physician Treat symptomatically.

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media Dry chemical
Special protective equipment for firefighters Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions Use personal protective equipment
Methods for cleaning up Soak up with inert absorbent material.

7. HANDLING AND STORAGE

Handling No special handling advice required
Storage Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection In case of insufficient ventilation wear suitable respiratory equipment
Hand protection Protective gloves
Eye protection Safety glasses with side-shields
Skin and body protection Lightweight protective clothing.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313

This product is not regulated by SARA.

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)

This product does not contain HAPs.

U.S. State Regulations

California Proposition 65

This product does not contain chemicals listed under Proposition 65

WHMIS hazard class:

Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

This material is sold for research and development purposes only. It is not for any human or animal therapeutic or clinical diagnostic use. It is not intended for food, drug, household, agricultural, or cosmetic use. An individual technically qualified to handle potentially hazardous chemicals must supervise the use of this material.

The above information was acquired by diligent search and/or investigation and the recommendations are based on prudent application of professional judgment. The information shall not be taken as being all inclusive and is to be used only as a guide. All materials and mixtures may be present unknown hazards and should be used with caution. Since Invitrogen Corporation cannot control the actual methods, volumes, or conditions of use, the Company shall not be held liable for any damages or losses resulting from the handling or from contact with the product as described herein. THE INFORMATION IN THIS MSDS DOES NOT CONSTITUTE A WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR ANY PARTICULAR PURPOSE.

End of Safety Data Sheet

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code Q10061MP A
Product name Qdot® 705 ITK™ streptavidin conjugate *2 µM solution*

Company/Undertaking Identification

INVITROGEN CORPORATON
5791 VAN ALLEN WAY
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
5250 MAINWAY DRIVE
BURLINGTON, ONT
CANADA L7L 6A4
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS**Hazardous/Non-hazardous Components**

The product contains no substances which at their given concentration, are considered to be hazardous to health. We recommend handling all chemicals with caution.

3. HAZARDS IDENTIFICATION**Emergency Overview**

The product contains no substances which at their given concentration, are considered to be hazardous to health

Form
Liquid

Principle Routes of Exposure/**Potential Health effects**

Eyes
Skin

No information available
No information available

3. HAZARDS IDENTIFICATION

Inhalation No information available
Ingestion No information available

Specific effects

Carcinogenic effects No information available
Mutagenic effects No information available
Reproductive toxicity No information available
Sensitization No information available

Target Organ Effects No information available

HMIS

Health	0
Flammability	0
Reactivity	0

4. FIRST AID MEASURES

Skin contact Wash off immediately with plenty of water
Eye contact Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion Never give anything by mouth to an unconscious person
Inhalation Move to fresh air
Notes to physician Treat symptomatically.

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media Dry chemical
Special protective equipment for firefighters Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions Use personal protective equipment
Methods for cleaning up Soak up with inert absorbent material.

7. HANDLING AND STORAGE

Handling No special handling advice required
Storage Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection In case of insufficient ventilation wear suitable respiratory equipment
Hand protection Protective gloves
Eye protection Safety glasses with side-shields
Skin and body protection Lightweight protective clothing.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313

This product is not regulated by SARA.

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)

This product does not contain HAPs.

U.S. State Regulations

California Proposition 65

This product does not contain chemicals listed under Proposition 65

WHMIS hazard class:

Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

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End of Safety Data Sheet

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code Q25021MP COMPONENT A
Product name Component A: Qtracker® 655

Company/Undertaking Identification

INVITROGEN CORPORATON
5791 VAN ALLEN WAY
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
5250 MAINWAY DRIVE
BURLINGTON, ONT
CANADA L7L 6A4
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS**Hazardous/Non-hazardous Components**

The product contains no substances which at their given concentration, are considered to be hazardous to health. We recommend handling all chemicals with caution.

3. HAZARDS IDENTIFICATION**Emergency Overview**

The product contains no substances which at their given concentration, are considered to be hazardous to health

Form
Suspension

Principle Routes of Exposure/**Potential Health effects**

Eyes
Skin

No information available
No information available

3. HAZARDS IDENTIFICATION

Inhalation No information available
Ingestion No information available

Specific effects

Carcinogenic effects No information available
Mutagenic effects No information available
Reproductive toxicity No information available
Sensitization No information available

Target Organ Effects No information available

HMIS

Health	0
Flammability	0
Reactivity	0

4. FIRST AID MEASURES

Skin contact Wash off immediately with plenty of water
Eye contact Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion Never give anything by mouth to an unconscious person
Inhalation Move to fresh air
Notes to physician Treat symptomatically.

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media Dry chemical
Special protective equipment for firefighters Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions Use personal protective equipment
Methods for cleaning up Soak up with inert absorbent material.

7. HANDLING AND STORAGE

Handling No special handling advice required
Storage Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection In case of insufficient ventilation wear suitable respiratory equipment
Hand protection Protective gloves
Eye protection Safety glasses with side-shields
Skin and body protection Lightweight protective clothing.

Hygiene measures
Environmental exposure
controls

Handle in accordance with good industrial hygiene and safety practice
Prevent product from entering drains.

9. PHYSICAL AND CHEMICAL PROPERTIES

General Information

Form Suspension

Important Health Safety and Environmental Information

Boiling point/range	°C No data available	°F No data available
Melting point/range	°C No data available	°F No data available
Flash point	°C No data available	°F No data available
Autoignition temperature	°C No data available	°F No data available
Oxidizing properties	No information available	
Water solubility	No data available	

10. STABILITY AND REACTIVITY

Stability	Stable.
Materials to avoid	No information available
Hazardous decomposition products	No information available
Polymerization	Hazardous polymerisation does not occur.

11. TOXICOLOGICAL INFORMATION

Acute toxicity

Principle Routes of Exposure/

Potential Health effects

Eyes	No information available
Skin	No information available
Inhalation	No information available
Ingestion	No information available

Specific effects

Carcinogenic effects	No information available
Mutagenic effects	No information available
Reproductive toxicity	No information available
Sensitization	No information available

Target Organ Effects

No information available

12. ECOLOGICAL INFORMATION

Ecotoxicity effects	No information available.
Mobility	No information available.
Biodegradation	Inherently biodegradable.
Bioaccumulation	Does not bioaccumulate.

13. DISPOSAL CONSIDERATIONS

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313

This product is not regulated by SARA.

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)

This product does not contain HAPs.

U.S. State Regulations

California Proposition 65

This product does not contain chemicals listed under Proposition 65

WHMIS hazard class:

Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

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End of Safety Data Sheet

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code Q25021MP COMPONENT B
Product name Component B: Qtracker® Carrier

Company/Undertaking Identification

INVITROGEN CORPORATON
5791 VAN ALLEN WAY
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
5250 MAINWAY DRIVE
BURLINGTON, ONT
CANADA L7L 6A4
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS**Hazardous/Non-hazardous Components**

The product contains no substances which at their given concentration, are considered to be hazardous to health. We recommend handling all chemicals with caution.

3. HAZARDS IDENTIFICATION**Emergency Overview**

The product contains no substances which at their given concentration, are considered to be hazardous to health

Form
Liquid

**Principle Routes of Exposure/
Potential Health effects**

Eyes
Skin

No information available
No information available

3. HAZARDS IDENTIFICATION

Inhalation No information available
Ingestion No information available

Specific effects

Carcinogenic effects No information available
Mutagenic effects No information available
Reproductive toxicity No information available
Sensitization No information available

Target Organ Effects No information available

HMIS

Health	0
Flammability	0
Reactivity	0

4. FIRST AID MEASURES

Skin contact Wash off immediately with plenty of water
Eye contact Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion Never give anything by mouth to an unconscious person
Inhalation Move to fresh air
Notes to physician Treat symptomatically.

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media Dry chemical
Special protective equipment for firefighters Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions Use personal protective equipment
Methods for cleaning up Soak up with inert absorbent material.

7. HANDLING AND STORAGE

Handling No special handling advice required
Storage Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection In case of insufficient ventilation wear suitable respiratory equipment
Hand protection Protective gloves
Eye protection Safety glasses with side-shields
Skin and body protection Lightweight protective clothing.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313

This product is not regulated by SARA.

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)

This product does not contain HAPs.

U.S. State Regulations

California Proposition 65

This product does not contain chemicals listed under Proposition 65

WHMIS hazard class:

Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

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The above information was acquired by diligent search and/or investigation and the recommendations are based on prudent application of professional judgment. The information shall not be taken as being all inclusive and is to be used only as a guide. All materials and mixtures may be present unknown hazards and should be used with caution. Since Invitrogen Corporation cannot control the actual methods, volumes, or conditions of use, the Company shall not be held liable for any damages or losses resulting from the handling or from contact with the product as described herein. THE INFORMATION IN THIS MSDS DOES NOT CONSTITUTE A WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR ANY PARTICULAR PURPOSE.

End of Safety Data Sheet

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code Q10001MP COMPONENT A
Product name Qdot® 605 ITK™ streptavidin conjugate *2 µM solution*

Company/Undertaking Identification

INVITROGEN CORPORATON
5791 VAN ALLEN WAY
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
5250 MAINWAY DRIVE
BURLINGTON, ONT
CANADA L7L 6A4
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS**Hazardous/Non-hazardous Components**

The product contains no substances which at their given concentration, are considered to be hazardous to health. We recommend handling all chemicals with caution.

3. HAZARDS IDENTIFICATION**Emergency Overview**

The product contains no substances which at their given concentration, are considered to be hazardous to health

Form
Liquid

Principle Routes of Exposure/**Potential Health effects**

Eyes	No information available
Skin	No information available

3. HAZARDS IDENTIFICATION

Inhalation No information available
Ingestion No information available

Specific effects

Carcinogenic effects No information available
Mutagenic effects No information available
Reproductive toxicity No information available
Sensitization No information available

Target Organ Effects No information available

HMS

Health	0
Flammability	0
Reactivity	0

4. FIRST AID MEASURES

Skin contact Wash off immediately with plenty of water
Eye contact Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion Never give anything by mouth to an unconscious person
Inhalation Move to fresh air
Notes to physician Treat symptomatically.

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media Dry chemical
Special protective equipment for firefighters Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions Use personal protective equipment
Methods for cleaning up Soak up with inert absorbent material.

7. HANDLING AND STORAGE

Handling No special handling advice required
Storage Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection In case of insufficient ventilation wear suitable respiratory equipment
Hand protection Protective gloves
Eye protection Safety glasses with side-shields
Skin and body protection Lightweight protective clothing.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313

This product is not regulated by SARA.

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)

This product does not contain HAPs.

U.S. State Regulations

California Proposition 65

This product does not contain chemicals listed under Proposition 65

WHMIS hazard class:

Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

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End of Safety Data Sheet

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

Product code A10196
Product name Qdot 625 streptavidin conjugate

Company/Undertaking Identification

INVITROGEN CORPORATON
1600 FARADAY AVENUE
PO BOX 6482
CARLSBAD, CA 92008
760-603-7200

INVITROGEN CORPORATION
2270 INDUSTRIAL STREET
BURLINGTON, ONT
CANADA L7P 1A1
800-263-6236

GIBCO PRODUCTS
INVITROGEN CORPORATION
3175 STALEY ROAD P.O. BOX 68
GRAND ISLAND, NY 14072
716-774-6700

2. COMPOSITION/INFORMATION ON INGREDIENTS**Hazardous/Non-hazardous Components**

The product contains no substances which at their given concentration, are considered to be hazardous to health

3. HAZARDS IDENTIFICATION**Emergency Overview**

The product contains no substances which at their given concentration, are considered to be hazardous to health

Form
Liquid

**Principle Routes of Exposure/
Potential Health effects**

Eyes	No information available
Skin	No information available
Inhalation	No information available

3. HAZARDS IDENTIFICATION

Ingestion No information available

Specific effects

Carcinogenic effects No information available

Mutagenic effects No information available

Reproductive toxicity No information available

Sensitization No information available

Target Organ Effects No information available

HMIS

Health	0
Flammability	0
Reactivity	0

4. FIRST AID MEASURES

Skin contact Wash off immediately with plenty of water
Eye contact Rinse thoroughly with plenty of water, also under the eyelids.
Ingestion Never give anything by mouth to an unconscious person
Inhalation Move to fresh air
Notes to physician Treat symptomatically.

5. FIRE-FIGHTING MEASURES

Suitable extinguishing media Dry chemical
Special protective equipment for firefighters Wear self-contained breathing apparatus and protective suit

6. ACCIDENTAL RELEASE MEASURES

Personal precautions Use personal protective equipment.
Methods for cleaning up Soak up with inert absorbent material.

7. HANDLING AND STORAGE

Handling No special handling advice required
Storage Keep in properly labelled containers

8. EXPOSURE CONTROLS / PERSONAL PROTECTION

Occupational exposure controls

Exposure limits

Engineering measures Ensure adequate ventilation, especially in confined areas

Personal protective equipment

Respiratory protection In case of insufficient ventilation wear suitable respiratory equipment
Hand protection Protective gloves
Eye protection Safety glasses with side-shields
Skin and body protection Lightweight protective clothing.
Hygiene measures Handle in accordance with good industrial hygiene and safety practice.

Environmental exposure controls Prevent product from entering drains.

9. PHYSICAL AND CHEMICAL PROPERTIES

General Information

Form Liquid

Important Health Safety and Environmental Information

Boiling point/range	°C No data available	°F No data available
Melting point/range	°C No data available	°F No data available
Flash point	°C No data available	°F No data available
Autoignition temperature	°C No data available	°F No data available
Oxidizing properties	No information available	
Water solubility	No data available	

10. STABILITY AND REACTIVITY

Stability	Stable.
Materials to avoid	No information available
Hazardous decomposition products	No information available
Polymerization	Hazardous polymerisation does not occur.

11. TOXICOLOGICAL INFORMATION

Acute toxicity

Principle Routes of Exposure/

Potential Health effects

Eyes	No information available
Skin	No information available
Inhalation	No information available
Ingestion	No information available

Specific effects

Carcinogenic effects	No information available
Mutagenic effects	No information available
Reproductive toxicity	No information available
Sensitization	No information available

Target Organ Effects No information available

12. ECOLOGICAL INFORMATION

Ecotoxicity effects	No information available.
Mobility	No information available.
Biodegradation	Inherently biodegradable.
Bioaccumulation	Does not bioaccumulate.

13. DISPOSAL CONSIDERATIONS

Dispose of in accordance with local regulations

14. TRANSPORT INFORMATION

IATA

Proper shipping name	Not classified as dangerous in the meaning of transport regulations
Hazard Class	No information available
Subsidiary Class	No information available
Packing group	No information available
UN-No	No information available

15. REGULATORY INFORMATION

International Inventories

U.S. Federal Regulations

SARA 313

This product is not regulated by SARA.

Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)

This product does not contain HAPs.

U.S. State Regulations

California Proposition 65

This product does not contain chemicals listed under Proposition 65

WHMIS hazard class:

Non-controlled

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

16. OTHER INFORMATION

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End of Safety Data Sheet



PbS Core EviDots in Toluene

MSDS

Section 1 - Chemical Product

Product Family #:	PbS Core EviDots
Substance:	Core EviDots packed in Toluene
Trade Names/Synonyms:	Core EviDots
Chemical Family:	Matrix: aromatic hydrocarbon Nanocrystal: IV-VI semiconductor compound

Section 2 - Composition, Information on Ingredients

Component	CAS#	EC#	% By Weight
Toluene	108-88-3	203-625-9	≥ 96
Lead Sulfide (as nanocrystal compound)	1314-87-0	215-246-6	≤ 4

Section 3 - Hazards Identification

Hazard Description:	Toxic, Dangerous to the Environment
NFPA Rating:	Health = 2, Fire = 3, Reactivity = 0

Emergency Overview	
Color:	Brown-Black
Physical Form:	Liquid
Odor:	Distinct aromatic odor
Major Health Hazards:	Respiratory tract irritation, skin irritation, eye irritation, aspiration hazard, central nervous system depression, nerve damage.
Physical Hazards:	Flammable

Potential Health Effects	
Inhalation:	Irritation, nausea, headache, drowsiness, dizziness, disorientation, sleep disturbances, loss of coordination, dilated pupils, kidney damage and liver damage
Skin Contact:	Irritation
Eye Contact:	Irritation
Ingestion:	Nausea, stomach pain, headache, drowsiness, dizziness, disorientation, sleep disturbances, loss of coordination, dilated pupils, kidney damage, liver damage, aspiration hazard.
Carcinogen Status:	Toluene
OSHA:	No
NTP:	No
IARC:	No

Section 4 - First Aid Measures

Inhalation:	If inhaled, remove to fresh air. If not breathing give artificial respiration and seek medical attention.
Skin Contact:	Wash skin with soap and water for at least 15 minutes while removing contaminated personal protective equipment, clothing and shoes. Seek medical attention if needed.
Eye Contact:	Irrigate eyes for at least 15 minutes. Seek medical attention.
Ingestion:	If ingested, do not induce vomiting, seek medical attention immediately.

Section 5 - Fire Fighting Measures

Extinguishing Media:	Dry chemical, carbon dioxide and foam extinguisher
Fire Fighting:	Avoid Inhalation of material or combustion by-products
Flash Point:	39F (4C) (closed cup)
Flammable Limits:	1.2% LEL -7.1% UEL
Autoignition Point:	896 F (480 C)
Flammability Class:	OSHA Class IB

Section 6 - Accidental Release Measures

Small Spills:	Absorb with spill pillow or other non-combustible material. Collect spilled material in appropriate container for disposal
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Section 7 - Handling and Storage

Store in a tightly closed container. Store in a cool dry place.

Section 8 - Engineering Controls & Personal Protective Equipment

Exposure Limits	
Toluene	200ppm OSHA TWA PEL 300ppm ceiling OSHA 50ppm ACGIH TWA (skin) 100ppm (375 mg/m ³) NIOSH TWA 10hour 190mg/m ³ DFG MAK 50ppm (191 mg/m ³) UK OES TWA

Ventilation:	Provide local exhaust ventilation system or work in a chemical fume hood. Considerations should be made for the use of non-sparking or intrinsically safe ventilation systems and equipment if explosive concentrations of material are present. Ensure compliance with applicable exposure limits.
Eye Protection:	Wear safety glasses with side shields as a minimum level of protection. If splash or splatter is possible, wear chemical/splash resistant safety goggles and or face shield. Emergency eye wash station and quick drench shower should be provided in the immediate work area as per the ANSI Z358.1 guidelines.
Clothing:	Wear appropriate chemical resistant clothing.
Gloves:	Wear appropriate chemical resistant gloves for type of exposure. (polyvinyl alcohol, Teflon™ and Viton™ are resistant to toluene exposure)
Respirator:	Refer to 29CFR1910.134 for selection of appropriate respiratory protection. Organic vapor cartridge with a ½ or full face mask, where toluene vapors do not exceed the assigned protection factor for the respirator. For unknown concentrations or IDLH atmospheres wear self-contained breathing apparatus or supplied air with escape bottle.

Section 9 - Physical & Chemical Properties

Lead Sulfide Core EviDots in Toluene	
Physical State:	liquid
Color:	Brown-Black
Odor:	distinct aromatic odor
Odor Threshold:	~10-15 ppm
Molecular Weight:	92.14 (toluene):MW of PbS EviDots N/A
Boiling Point:	232F (111 C)
Freezing Point:	-139 F (-95 C)
Vapor Pressure:	22mmHg @20C
Vapor Density (air = 1):	3.14
Specific Gravity (water = 1):	0.8669
pH:	No data available

Section 10 - Reactivity

Stability:	Stable at standard temperatures and pressure.
Conditions to avoid:	Avoid heat, sparks and other sources of ignition.
Incompatible:	Incompatible with oxidizing materials, halogens, acids, combustible materials and metal salts.
Hazards Decomposition:	Combustion produces toxic by-products.
Polymerization:	Will not polymerize.

Section 11 - Toxicological Information

Toluene	
Irritation Data:	300 ppm eyes-human; 435 mg skin-rabbit mild; 500 mg skin-rabbit moderate; 20mg/24hours skin-rabbit moderate
Toxicity Data:	719ul/kg oral-man LDLo; 50 mg/kg oral-human LDLo; 200 ppm inhalation-human TCLo; 100 ppm inhalation-man TCLo; 636 mg/kg oral-rat LD50; 49 gm/m ³ /4 hours inhalation-rat LC50; 1332 mg/kg intraperitoneal-rat LD50; 1960 mg/kg intravenous-rat LD50; 6900 mg/kg unreported-rat LD50; 400 ppm/24hours inhalation-mouse LC50; 59 mg/kg intraperitoneal-mouse LD50; 2250 mg/kg subcutaneous-mouse LD50; 2gm/kg unreported-mouse LD50; 14100ul/kg skin-rabbit LD50; 130mg/kg intravenous-rabbit LDLo; 1600 ppm inhalation-guinea pig LCLo
Local Effects:	Irritant; inhalation, skin, eye
Slightly Toxic:	Inhalation & dermal absorption.
Moderately Toxic:	Ingestion
Target Organs:	Nervous system
Additional Toxicological Information:	To the best of our knowledge the acute and chronic toxicity of this substance is not fully known. Zinc Selenide, in the form of a nanocrystal may or may not present the same health hazards as a larger zinc or selenium containing molecules. It is therefore encouraged to use caution when handling this product as its toxicity and modes of exposure are not well characterized or understood.

Section 12 - Ecological Information

Do not allow material to be released to the environment (ground, air or water bodies) without proper permits. Relatively non-persistent in the environment. Accumulates very little in the bodies of living organisms.

Section 13 - Transportation Information

U.S. DOT:	Class 3, packing group II, UN1294
Canadian Transportation of Dangerous Goods:	UN 1294 Class 3
Land Transport ADR/RID:	UN1294, Class 3, Class Code F1, Pack group II
Air Transport IATA/ICAO:	UN1294, Class or Division 3, pack group II
Exceptions:	49 CFR 173.4

Section 14 - Disposal

U.S. EPA 40 CFR 262: Hazardous Waste Number: D001 (Flammable), U220 (toluene)
Dispose in accordance with all applicable local, state and federal regulations.

Section 15 - Regulatory Information

US Regulations

Toluene
CERCLA: 1000 Lbs RQ
SARA Title III, sec. 302, 304: Not regulated
SARA Title III, Section 311/312
Acute: Yes
Chronic: Yes
Fire: No
Reactive: No
Sudden Release: No
US Inventory (TSCA) listed: Yes

Canadian Regulations

WHMIS Classification: Not available

European Regulations

EC Classification:
F Highly Flammable
Xn-Harmful

EC Risk Phrases

R11, R20, S2, S16, S25, S29, S33

Section 16 - Other Information

The above information is believed to be correct but does not purport to be all inclusive and shall be used only as a guide. Evident Technologies shall not be held liable for any damage resulting from handling or from contact with the above product. See packing slip for additional terms and conditions of sale.

Revision Date: 9/16/2008
Creation Date: 12/7/2004



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REFERENCES

References for Published Research on Quantum Dots:

K. Chen, J. Kibsgaard, and T. Jaramillo, "Nanostructuring MoS₂ for Photoelectrochemical Water Splitting," *Solar Hydrogen and Nanotechnology*, 2010, **7770**, 77700K-1-7.

K. Zhang, Y. Osakada, M. Vrljic, L. Chen, H.V. Mudrakola, and B. Cui, "Single-molecule imaging of NGF axonal transport in microfluidic devices," *Lab Chip*, 2010, **10**, 2566-2573.

M.A Thompson, M. D. Lew, M. Badieirostami, and W. E. Moerner, "Localizing and Tracking Single Nanoscale Emitters in Three Dimensions with High Spatiotemporal Resolution Using a Double-Helix Point Spread Function" *Nano Letters*, 2010, **10**, 211-218.

EXPERIMENTAL PROTOCOL INFORMATION FOR THE USE OF INVITROGEN PRODUCTS

Qdot® Streptavidin Conjugates

Table 1. Contents and storage information.

Material	Amount	Concentration	Storage	Stability
Qdot® streptavidin conjugate	200 µL or 50 µL	1 µM solution in 1 M betaine, 50 mM borate, pH 8.3 with 0.05% sodium azide*	<ul style="list-style-type: none"> • 2–6°C • Do not freeze 	When stored as directed, product is stable for at least 6 months.

*Betaine acts as a cryoprotectant during shipping and does not affect the fluorescence of Qdot® conjugates.

Approximate fluorescence excitation/emission maxima: See Figure 3.

Introduction

Structure of Qdot® Nanocrystals

The Qdot® streptavidin conjugate is made from a nanometer-scale crystal of a semiconductor material (CdSe), which is coated with an additional semiconductor shell (ZnS) to improve the optical properties of the material. These materials have a narrow, symmetric emission spectrum with the emission maximum near 525 nm (Q10141MP), 565 nm (Q10131MP), 585 nm (Q10111MP), 605 nm (Q10101MP), 625 nm (A10196), 655 nm (Q10121MP), 705 nm (Q10161MP), or 800 nm (Q10171MP). The Qdot® 705 and Qdot® 800 streptavidin conjugates, which include CdSeTe, are made in a similar fashion. This core-shell material (Figure 1A) is further coated with a polymer shell that allows the materials to be conjugated to biological molecules and to retain their optical properties. This polymer shell is directly coupled to streptavidin (Figure 1B). The Qdot® streptavidin conjugate is the size of a large macromolecule or protein (~15–20 nm).

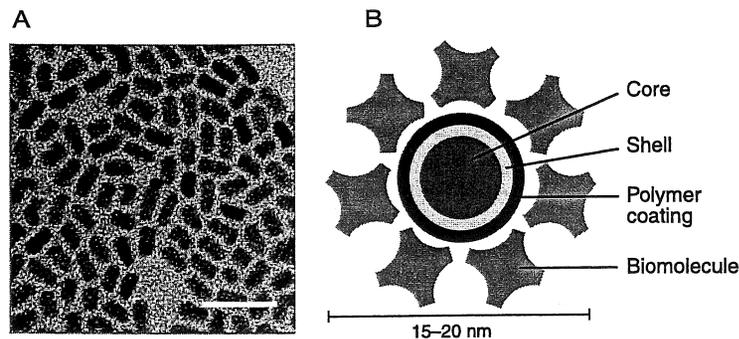


Figure 1. **A.** Transmission electron microscope image of core-shell Qdot® nanoparticles at 200,000x magnification. Scale bar = 20 nm. **B.** Schematic of the overall structure of a Qdot® streptavidin conjugate. The layers represent the distinct structural elements of the Qdot® nanocrystal conjugates, and are roughly to scale.

Optical Properties

The optical properties of Qdot® conjugates are different than those of typical dye molecules. The color of light that the Qdot® nanocrystal emits is strongly dependent on the particle size, creating a common platform of labels from the green to the red, all manufactured from the same underlying semiconductor material (see *Bibliography*, references 1–11 in the *Appendix*). The size of Qdot® nanocrystals are tightly controlled in the production process, resulting in materials with narrow and symmetric emission spectra, that are extremely bright and photostable. While the fluorescence emission from the Qdot® 705 and Qdot® 800 streptavidin conjugates are broader than the other Qdot® conjugates, the fluorophores have similar intensities and photostabilities. Note that the 705 and 800 nm quantum dot emission cannot be seen by eye, but is easily detected by many cameras and detectors. These properties are exploited in a variety of immunofluorescence techniques, and can result in substantially better results than are attainable with conventional immunofluorescent labels (see *Bibliography*, references 12–18). Though these materials are compatible with a number of standard immunofluorescent techniques, there are some novel aspects of their chemistry and detection that require careful consideration to attain optimal assay results.

Biological Activity

The surface chemistry dictates many of the important properties of the Qdot® nanocrystal in a biological experiment. The surface has been prepared to have a low nonspecific signal when incubated with samples in a variety of aqueous buffers. Qdot® nanocrystals have been coupled to streptavidin directly through an active ester coupling reaction.¹⁹ This yields a material with streptavidin covalently attached on the surface (typically 5–10 streptavidins/Qdot® conjugate), which results in Qdot® streptavidin conjugates with high specific biological activity. The probes should generally be used as if there were one streptavidin per Qdot® nanocrystal. Though one quantum dot is capable of bridging multiple antigens through a biotinylated IgG, the dominant binding mode is one Qdot® conjugate per analyte if the assay is carried out at a saturating concentration. (Figure 2.)

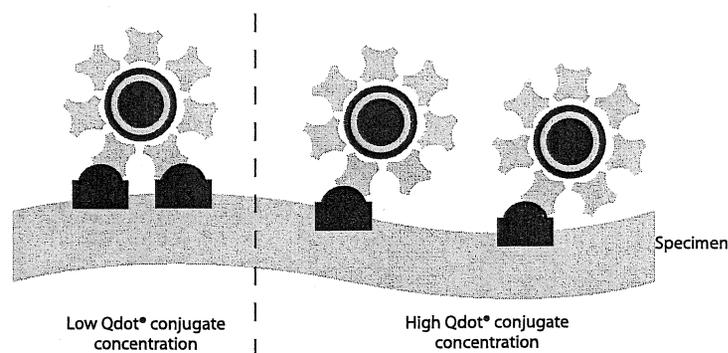


Figure 2. Impact of working at the appropriate concentration range for Qdot® streptavidin conjugates. Due to the multivalency of the conjugates, use of conditions below the appropriate saturation concentration may result in artificially reduced signals due to antigen bridging with a single quantum dot conjugate.

Spectral Characteristics

Typical fluorescent dyes have excitation and emission spectra with a relatively small Stokes shift, which means that the optimal excitation wavelength is close to the emission peak. Filter sets used with fluorescent dyes reflect this characteristic.²⁰ Qdot® nanocrystals have absorbance spectra that increase dramatically to the blue of the emission (Figure 3). These unique spectral properties are due to the semiconductor that makes up the core of the Qdot® conjugates, which gives rise to both the absorbance and emission properties of the materials (see *Bibliography*, references 1–11). Despite the broad absorbance, the emission wavelength is independent of the excitation wavelength; so whether exciting at 633 nm or at 400 nm, the shape of the emission remains the same, while the intensity is approximately 11-fold

higher with 400 nm excitation. The absorbance and excitation at shorter wavelength, with fixed emission for the material results in a large “apparent Stokes shift” which improves sensitivity by reducing auto-fluorescence, and greatly simplifies the multiplexed detection of several Qdot® conjugates. See *Appendix 3* for extinction coefficients at common excitation wavelengths of the different materials.

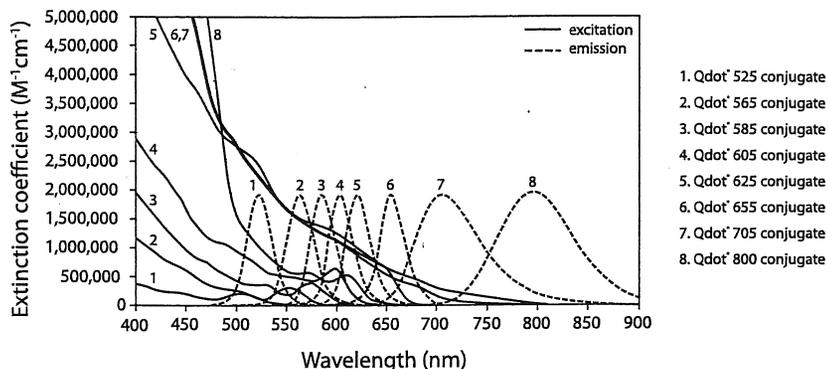


Figure 3. Typical absorption and emission spectra of Qdot® 525 streptavidin conjugate (1), Qdot® 565 streptavidin conjugate (2), Qdot® 585 streptavidin conjugate (3), Qdot® 605 streptavidin conjugate (4), Qdot® 625 streptavidin conjugate (5), Qdot® 655 streptavidin conjugate (6), Qdot® 705 streptavidin conjugate (7), Qdot® 800 streptavidin conjugate (8).

Optical Filter Selection

To achieve the optimal signal from Qdot® streptavidin conjugates, we recommend using Qdot® optimized filter sets that are available from Omega Optical, Semrock, or Chroma Technology Corporation (see *Appendix 2* for details).

The Qdot® streptavidin conjugate can also be viewed through some standard filter sets,

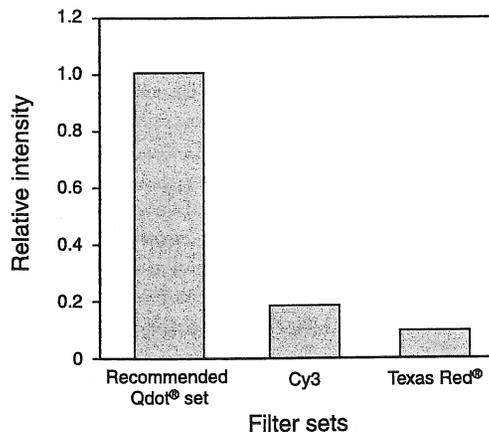


Figure 4. Detection of Qdot® conjugates on tissue sections with recommended and standard filter sets. Mouse kidney sections were stained with Qdot® 605 streptavidin conjugate, and then images were collected on a Nikon epi-fluorescence microscope in 16 bit capture mode. The mean fluorescence of positively stained samples was extracted using Scion Image software. The recommended Qdot® filter set included a 460 nm short pass exciter, a 475 nm dichroic, and a 605/20 nm band pass emitter. The Cy3 filter set included a 545/30 nm exciter, a 570 nm dichroic, and a 610/75 nm emitter. The Texas Red® filter set included a 560/40 nm exciter, a 595 nm dichroic, and a 630/60 nm emitter.

albeit with lower detection efficiency and reduced brightness. For example, three Omega Optical standard filter sets capable of detecting Qdot® 705 conjugates are XF140-2 (Alexa Fluor® 633 & Alexa Fluor® 647), XF70 (Alexa Fluor® 660 & Cy5), and XF141-2 (Cy5.5). Visualization of Qdot® conjugates using a custom filter set is preferred because excitation and detection is less efficient using filters that have not been selected specifically for use with Qdot® conjugates. Using a custom filter set, Qdot® 605 streptavidin conjugate signal is approximately five times as bright as it is using the TRITC filter set, and approximately ten times brighter than it is using the Texas Red®/Cy3.5 filter set (Figure 4). Qdot® optimal filters and standard filter sets are available from many different filter manufacturers. *Appendix 2* illustrates some common filter sets and the optimal filter set recommendations for the available Qdot® streptavidin conjugates. Use of the optimal filter set is critical for attaining optimal signal and sensitivity in your experiments. For detailed technical notes and examples of how to set up specific instruments to detect Qdot® conjugates optimally, visit www.invitrogen.com.

Before You Begin

Qdot® nanocrystals have chemical and optical properties that provide significant advantages over conventional fluorophores in both sensitivity and stability in immunofluorescent labeling of cells and tissue sections. We have conjugated streptavidin to Qdot® nanocrystals to allow use of the materials in a wide variety of labeling applications. Biotinylated antibodies can be obtained or easily prepared using available reagents without substantial disruption of the specificity or affinity of the native antibody. These antibodies can be specifically bound to the biological specimen and then detected through a second round of staining with a Qdot® streptavidin conjugate.

Note: The 705 and 800 nm emission cannot be seen by eye, but is easily detected by many cameras and detectors.

General Considerations

Buffer compatibility

The Qdot® streptavidin conjugates have stable emission in a number of distinct buffers, across a range of pH conditions. At working concentrations, the quantum yield and colloidal dispersion of these materials has been found to be remarkably stable across pH 6–9 (not investigated outside this range) in Tris, HEPES, phosphate, and borate buffers. The Qdot® streptavidin conjugates are stable and non-aggregated in buffered NaCl up to 200 mM at working concentrations. Higher salt concentrations result in microscopic aggregates, but do not appear to cause bulk precipitation of the materials at working dilutions. In addition, a number of surfactants and additives such as Tween 20, Triton® X-100, Pluronic F68, NDSB 201, and EDTA, among others have been shown to maintain the fluorescence when used at 0.05% concentration. In contrast, gelatin and dextran sulfate were found to promote aggregation of the Qdot® 605 streptavidin conjugate at 0.05% concentration, and should be avoided in labeling applications. In general, we recommend storage of Qdot® conjugates at the concentration at which it is shipped, rather than at a high dilution. Storage of Qdot® conjugates at working dilution over longer periods of time may result in substantial performance degradation. While we have not characterized the stability of all Qdot® streptavidin conjugates in all of these conditions, we anticipate similar levels of stability across the range of product colors.

Controls

If you are using the Qdot® conjugates or the labeling protocol for the first time, we recommend including a positive labeling control. For example, an optional control for Qdot® goat-anti-mouse secondary antibodies is anti-OxPhos Complex V Inhibitor Protein (Invitrogen Cat. no. A21355) which targets mitochondria. Other antibodies used in the optimization of this protocol and suitable as positive controls include: rabbit-anti-giantin

(Covance, Cat. no. PRB-114C), rabbit anti-AIF (Cell Signaling, Cat. no. 4642), mouse anti-ki-67 (Ventana Medical Systems, Cat. no. 790-2910), mouse anti-alpha tubulin (Sigma, Cat. no. T6074), rabbit anti-alpha tubulin (Affinity Bioreagents, Cat. no. PA1-20988), mouse anti-nucleosome (BD Pharmingen, Cat. no. 51-80591N), and mouse anti-nucleolin (Invitrogen, Cat. no. 39-6400; Stressgen, Cat. no. KAM-CP100).

Antigen labeling with Qdot® conjugates

Detecting cellular targets with Qdot® streptavidin conjugates can be performed individually by using a single Qdot® streptavidin conjugate with one primary antibody, or multiplexed by using a combination of primary antibodies and various Qdot® nanocrystal colors. Golgi, tubulin, mitochondrial, peroxisome, nucleolin, nucleosome, and ki-67 targets have been validated with this labeling protocol (for a complete list of primary antibodies validated, see above). Other targets and cell lines, however, may require further optimization of this protocol. For example, reducing the fixation time may improve cell penetration and conjugate access for some targets as may increasing the concentration of permeabilization reagent or incubation time with the permeabilization buffer.

Qdot® nanocrystal toxicity

We have not investigated the toxicity of the Qdot® streptavidin conjugate. The materials are provided in a solution which is ~2 mM total Cd concentration; however, the CdSe core is encapsulated in a shell of ZnS and the polymer shell, which may help prevent formation of free Cd. We have demonstrated the utility of these materials in a variety of live-cell *in vitro* labeling experiments, but do not have systematic data on the toxicity of the materials to humans, to animals, or to cells in culture.

FRET or close-proximity quenching

We have not systematically investigated the energy transfer properties of the Qdot® nanocrystals, though they may have useful properties as energy transfer donors and acceptors. We have investigated the fluorescence of Qdot® 605 streptavidin conjugates which are coupled to each other through a bis-biotin linker, and found that the emission intensity of the materials was unperturbed at any concentration of biotin cross-linker. These results suggest that the interparticle quenching of these Qdot® conjugates is negligible. Recent published literature indicates that Qdot® nanocrystals can be used as energy acceptors in time-resolved FRET (TR-FRET) studies.²¹

Disposal of Qdot® Conjugate

The Qdot® conjugate contains cadmium and selenium in an inorganic crystalline form. Dispose of the material in compliance with all applicable local, state, and federal regulations for disposal of these classes of material. For more information on the composition of these materials, consult the Material Safety Data Sheet.

Materials Required but Not Provided

- 10X PBS, pH 7.4 (phosphate buffered saline, Invitrogen Cat. no. 70011-044)
- Fixative: 4% formaldehyde in PBS
- Permeabilization buffer: 0.25% Triton® X-100 in PBS
- Wash buffer: 1X PBS, pH 7.4
- Endogenous Biotin Blocking Kit (Invitrogen Cat. no. E21390)
- Blocking buffer: 6% BSA (bovine serum albumin) and 10% normal serum in PBS
- Primary antibody, biotinylated primary antibody, or other biotinylated protein
- Biotinylated secondary antibody if using non-biotinylated primary antibody
- Secondary incubation buffer: 6% BSA in PBS
- Dehydration solutions: Prepare in containers suitable for a dehydration series ethanol/water dilutions (v/v) of 30%, 50%, 70%, and 90%, as well as 100% ethanol and 100% toluene
- Mounting reagent: Cytoseal™ 60 (Richard-Allan Scientific, Cat. no. 8310-16) is recommended

Preparing Solutions

The amount of solutions prepared as described below is sufficient to process approximately 20 coverslips.

Fixative: 4% formaldehyde in PBS

Prepare 40 mL of fixative **fresh** by combining 10 mL of formaldehyde (ultrapure, methanol-free, 16% formaldehyde solution, Polysciences, Inc. Cat. no. 18814) and 4 mL of 10X PBS, pH 7.4 (Invitrogen Cat. no. 70011-044) with 26 mL of distilled water. Mix well.

Permeabilization buffer: 0.25% Triton® X-100 in PBS

Prepare 40 mL of Permeabilization buffer by adding 100 µL of Triton® X-100 (Sigma, Cat. no. T9284) to 40 mL of 1X PBS. Stir until the Triton® X-100 goes into solution. Store any remaining Permeabilization buffer at 4°C for up to a week.

Wash buffer: 1X PBS, pH 7.4

Prepare 2 L of 1X PBS by combining 200 mL of 10X PBS, pH 7.4 (Invitrogen Cat. no. 70011-044) and 1.8 L of distilled water. Mix well.

Blocking buffer: 6% BSA/10% normal serum in PBS

To prepare 50 mL Blocking buffer, add 3 g of BSA (RIA grade, Fraction V, minimum 96%, Sigma Cat. no. A-7888), 5 mL of normal serum from the host species of the secondary antibody (preferably heat inactivated at 56°C for 1 hour), and 5 mL of 10X PBS, pH 7.4 to 40 mL distilled water. Mix well until the BSA is completely dissolved and adjust the volume to 50 mL with distilled water. Mix well with gentle rocking or stirring. If storing the blocking buffer, add sodium azide to a final concentration of 0.02% and store at 4°C.

Note: Avoid using blocking buffers with casein as casein can cause quenching of Qdot® conjugates.

Primary antibody, biotinylated primary antibody, or other biotinylated protein

Dilute the primary antibody, biotinylated primary antibody, or other biotinylated protein in Blocking buffer at the concentration recommended by the manufacturer.

Note: Briefly centrifuge the primary antibody prior to use. You may need to titrate the primary antibody concentration in preliminary experiments to achieve optimal target labeling.

Biotinylated secondary antibody (required if using non-biotinylated primary antibody)

Dilute the biotinylated secondary antibody in Blocking buffer at the concentration recommended by the manufacturer.

Secondary incubation buffer: 6% BSA in PBS

To prepare 50 mL Secondary incubation buffer, add 3 g of BSA (RIA grade, Fraction V, minimum 96%, Sigma Cat. no. A-7888) and 5 mL of 10X PBS, pH 7.4 to 40 mL of distilled water. Mix well until BSA is completely dissolved and adjust the final volume to 50 mL with distilled water. Mix well with gentle rocking or stirring. If storing the Secondary incubation buffer, add sodium azide to a final concentration of 0.02% and store at 4°C.

Dehydration solutions

Prepare in containers suitable for a dehydration series with ethanol/water dilutions (v/v) of 30%, 50%, 70%, and 90%, as well as 100% ethanol and 100% toluene.

Preparing Qdot® Streptavidin Conjugate

Do not vortex the Qdot® streptavidin conjugate vial. Prepare the required amount of the diluted Qdot® streptavidin conjugate needed for the experiment on the day of use. You will need 40–200 µL Qdot® streptavidin conjugate per coverslip depending on your protocol and the type of humidity chamber you use.

- 1.1 Centrifuge the Qdot® streptavidin conjugate vial at 5000 × g for 3 minutes prior to use. Use only the supernatant and discard any pellet.

- 1.2 Dilute the conjugate by adding 2 μL of the stock (1 μM) conjugate to 100 μL Secondary incubation buffer immediately prior to use to obtain the Qdot[®] streptavidin conjugate concentration of 20 nM (you may use between 10 nM and 40 nM Qdot[®] streptavidin conjugate final concentration).
- 1.3 Use the diluted Qdot[®] streptavidin conjugate immediately for the current experiment. **Do not** store any diluted Qdot[®] streptavidin conjugate.

Preparing Cells

Culture cells in the appropriate medium to the desired confluency and physiological state (typically 1–2 days for HeLa cells). Make sure the cells are below 80% confluency at the time of fixation, depending on experimental requirements and the imaging method.

Experimental Protocols

Cell Labeling Protocol

Please read entire protocol before starting.

This protocol was validated with HeLa and NIH 3T3 cells, but can be adapted for any adherent mammalian cell types. For tissue labeling protocols, refer to the Qdot[®] Conjugates Protocol Handbook (mp19029) available for downloading from www.invitrogen.com. Perform all steps of the procedure at room temperature. For a flowchart of the labeling protocol, see Figure 5.

The optimal signal-to-background ratio is obtained from an assay that has been optimized with respect to all of the probe concentrations. Excess concentrations of the primary or secondary antibodies can cause increases in background staining. The optimal concentrations of these reagents can be determined by testing the signal-to-background ratio of control and positive samples that are treated with a dilution series of each antibody, and then stained under typical conditions.

Fixation and Permeabilization

- 2.1 Rinse cells briefly in 1X PBS prewarmed to 37°C.
- 2.2 Fix cells in Fixative for 15 minutes. Pour off or aspirate the solution.
- 2.3 Wash cells 3 times with 1X PBS. If cells are grown on coverslips in a large dish (i.e., 100 mm petri dish), add sufficient volume of 1X PBS to completely cover the specimen (~5 mL for a 100 mm petri dish), swirl gently, pour off or aspirate the solution and repeat. If cells are grown on coverslips in a smaller container (i.e., 6-well plate), wash cells 3 times for 5 minutes each by adding enough 1X PBS to completely cover the specimen. After 5 minutes, pour off or aspirate the solution.
- 2.4 Permeabilize the specimen with Permeabilization buffer for 15 minutes. Pour off or aspirate the solution.
- 2.5 Repeat step 2.3.

Note: Changing fixation or permeabilization times, or reagent concentrations may be needed for achieving labeling of certain targets.

Target Labeling and Detection

All incubations are performed in a humidity chamber at room temperature (see **Note**, below). Avoid specimen drying as this can cause high levels of non-specific background and autofluorescence. At the end of each step, carefully remove or blot excess solution from the sample before moving to the next step.

Note: A simple humidity chamber prevents labeling reagent concentration changes due to

evaporation during incubations. To make a simple humidity chamber, place a piece of filter paper in a Petri dish and saturate the filter paper with water. Place a piece of laboratory film on the filter paper and place the coverslip with the solution on the laboratory film. During incubations, place a lid on the Petri dish. Gentle agitation during incubation steps is optional.

2.6 Perform blocking of endogenous biotin with the Endogenous Biotin Blocking Kit (Invitrogen, Cat. no. E21390) as follows. The kit contains two reagents supplied in a ready-to-use format.

a) Incubate in Component A for 30 minutes.

b) Wash 3 times with 1X PBS for 5 minutes each.

c) Incubate in Component B for 30 minutes.

d) Wash 3 times with 1X PBS for 5 minutes each.

2.7 Add Blocking buffer and incubate for 1 hour. Pour off or aspirate the blocking buffer.

2.8 Incubate for 1 hour with primary antibody, biotinylated primary antibody, or other biotinylated protein, diluted in blocking buffer. Pour off or aspirate the solution.

2.9 Wash 3 times with 1X PBS for 5 minutes each.

Note: If using a biotinylated secondary antibody, repeat step 2.8 with the biotinylated antibody and perform wash step 2.9 before proceeding to the next step.

2.10 Incubate for 1 hour with Qdot® streptavidin conjugate, diluted to an optimal concentration (titrate between 10 nM and 40 nM for optimal results) in secondary incubation buffer. Pour off or aspirate the solution.

2.11 Wash 3 times with 1X PBS for 5 minutes each.

2.12 **Optional:** If counterstaining is necessary, most counterstain procedures may be performed at this point, followed by necessary wash steps.

Note: UV excitation of DAPI (Invitrogen, Cat. no. D1306) resulting in emission around 600 nm has been observed, and may not be appropriate for use with all Qdot® nanocrystals. Several other nuclear counterstain options are available in a kit form with optimized protocols for use with Qdot® conjugates such as SelectFX® Nuclear Labeling Kit (Invitrogen, Cat. no. S33025), SYTOX® Green nucleic acid stain (Invitrogen, Cat. no. S7020), 7-aminoactinomycin D (7-AAD) (Invitrogen, Cat. no. A1310), propidium iodide (Invitrogen, Cat. no. P3566), and TO-PRO®-3 iodide (Invitrogen, Cat. no. T3605).

2.13 Dehydrate the specimen by submerging the coverslip sequentially for 5 seconds in 30%, 50%, 70%, and 90% ethanol/water, twice in 100% ethanol, once in 100% toluene, and the final dip in 100% toluene for 10 seconds.

When transferring the coverslip from the last toluene wash to the mounting medium, blot any excess toluene with a laboratory wipe, but allow a sheen of toluene to remain on the coverslip surface. **Do not** allow the residual toluene on the coverslip to completely evaporate prior to mounting, as this can lead to cell morphology damage and/or high background.

2.14 Mount using one drop of Cytoseal™ 60 and allow the specimen to cure at least 4 hours before imaging. For optimal performance, image within 24 hours.

Imaging Guidelines

For optimal imaging of Qdot® streptavidin conjugates, including reduced spectral bleedthrough effects in multi-color applications, use filter sets optimized for the excitation and emission of the Qdot® conjugates in use. These filters are available from Omega Optical, Semrock, or Chroma Technology Corporation (see *Appendix 2* for details). For additional information, visit probes.invitrogen.com/products/qdot.

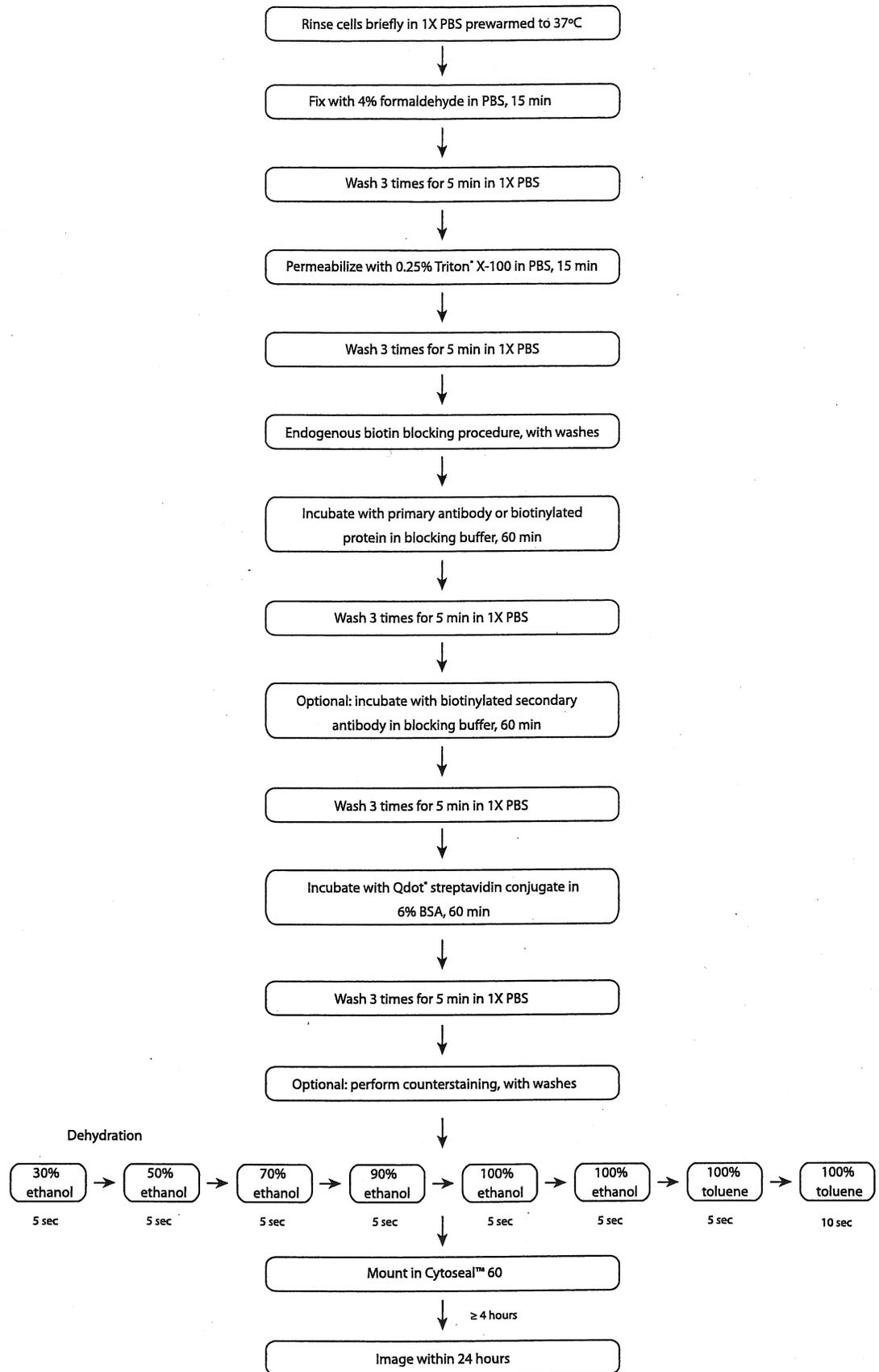


Figure 5. Cell labeling flowchart.

Appendix 1: Troubleshooting Guide

The properties of Qdot® conjugates are different from fluorescent dyes and may require slight modifications to current protocols. We've included this section to help with some specific issues that may arise while using these materials.

No Signal

Setup suitability

Make sure that you are using an appropriate filter set to detect the signals. See *Appendix 2* for a list of appropriate and optimal filters for the Qdot® conjugates. Contact Technical Support (probestech@invitrogen.com) for more details on particular filter set requirements.

Qdot® conjugate luminosity

Qdot® conjugates normally fluoresce brightly under a hand-held ultraviolet lamp (long wave, such as the type used to visualize ethidium bromide on agarose gels). The 705 and 800 nm quantum dot emission cannot be seen by eye, but is detected by many cameras and detectors. Though we have not seen pronounced loss of fluorescence of these materials under any storage conditions that we have investigated, we have not been able to examine all storage conditions. If the Qdot® streptavidin conjugates do not appear to fluoresce under the long wave UV excitation, contact Technical Support (probestech@invitrogen.com) for assistance.

Specificity and titer of primary antibody

Make sure the antibody recognizes the intended targets and that there is sufficient primary antibody bound to the targets. This verification can be performed by ELISA based capture of the antigen of interest, or by other techniques that can be found in lab manuals such as the *Current Protocols in Immunology*.²²

Biotinylation of antibody

Make sure your antibodies (the primary antibodies for two-layer and the secondary antibodies for three-layer detection) are effectively biotinylated. It may be necessary to independently adjust the concentration of the primary and secondary antibodies used in the assay to obtain optimal signal and minimal background.

High Background

High level of endogenous biotin

Increasing blocking times for the Endogenous Biotin Blocking Kit (Invitrogen Cat. no. E21390) steps may help reduce background due to endogenous biotin.

BSA lot variability

BSA used in the blocking buffer can vary by lot and producer. If unacceptable background is observed, re-optimization of blocking conditions may be required for best results when substituting alternate sources for BSA.

Antibody and Qdot® conjugate concentration optimization

Adjusting the level of biotinylated antibody for the staining can often be used to optimize the specific signal. High levels of biotinylated antibody maybe necessary to obtain the specific labeling, but overly high levels contribute to the nonspecific binding of the antibody to the sample. Nonspecifically bound biotinylated antibody will bind to the Qdot® streptavidin conjugate, resulting in higher background staining.

Appendix 2: Optimal Usable Filter Sets for Qdot® Conjugates

Table 2. Omega Optical filter set for Qdot® streptavidin conjugates.

Color	Optimal filter sets	Usable filter sets
525	XF301 Qdot®525 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP Emitter: 525WB20)	XF100-3, XF100-2, XF115-2, XF89-2
565	XF302 Qdot® 565 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter 565WB20)	XF104-2, XF105-2
585	XF303 Qdot® 585 filter set (Exciter: 1 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 585WB20)	XF101-2, XF137-2, XF152-2
605	XF304 Qdot® 605 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 605WB20)	XF108-2, XF102-2, XF103-2
655	XF305 Qdot® 655 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 655WB20)	XF102-2, XF40-2, XF42, XF45
705*	XF306 Qdot® 705 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 710AF40)	XF140-2, XF70, XF110-2, XF141-2, XF48-2
800 *	XF307 Qdot® 800 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 800WB80)	XF308 Qdot® 800 filter set for multiplexing (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 840WB80)
All colorst	XF300 Qdot® filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP Emitters: 800WB80, 840WB80, 710AF40, 655WB20, 605WB20, 585WB20, 565WB20, and 525WB20)	XF129-2, XF130-2

*The 705 and 800 nm quantum dot emission cannot be seen by eye and must be detected with an IR-sensitive detector.

†For viewing multiple colors of Qdot® nanocrystals through microscope eyepieces.

Table 3. Semrock filter sets for Qdot® streptavidin conjugates.

Color	Optimal filter sets	Usable filter sets
525	Brightline® QD525-A Filter Sets: QD525-A-000 or QD525-A-000-ZERO (Exciter: FF01-435/40-25) (Dichroic: FF510-Di01-25 x 36) (Emitter: FF01-525/15-25)	GFP-3035B
565	--	FITC-3504B or YFP-2427A
585	--	TRITC-A
605	Brightline® QD605-A Filter Sets: QD605-A-000 or QD605-A-000-ZERO (Exciter: FF01-435/40-25) (Dichroic: FF510-Di01-25 x 36) (Emitter: FF01-605/15-25)	TRITC-A
625	Brightline® QD625-A Filter Sets: QD625-A-000 or QD625-A-000-ZERO (Exciter: FF01-435/40-25) (Dichroic: FF510-Di01-25 x 36) (Emitter: FF01-625/15-25)	Texas Red® (4040B)
655	Brightline® QD655-A Filter Sets: QD655-A-000 or QD655-A-000-ZERO (Exciter: FF01-435/40-25) (Dichroic: FF510-Di01-25 x 36) (Emitter: FF01-655/15-25)	Texas Red® (4040B)
705*	--	Cy5-4040A or Cy5.5-A
800*	--	Cy7-A
LP multi†	QDLP-A Filter Set: QDLP-A-000 (Exciter: FF01-435/40-25) (Dichroic: FF510-Di01-25 x 36) (Emitter: FF01-500/LP-25)	CFW-LP01-CLINICAL

*The 705 and 800 nm quantum dot emission cannot be seen by eye and must be detected with an IR-sensitive detector.
†For viewing multiple colors of Qdot® nanocrystals through microscope eyepieces.

Table 4. Chroma Technology filter sets for Qdot® streptavidin conjugates.

Color	Optimal filter sets	Usable filter sets
525	Qdot® 525 filter set (20 nm EM; 32006) (460SPUV/475DCXRU/D525/20nm) Qdot® 525 filter set (40 nm EM; 32010) (460SPUV/475DCXRU/D525/40nm)	FITC/RSGFP/Bodipy®/Fluo-3/DiO (41001), FITC/RSGFP Longpass (40012), BFP to GFP FRET (31032), BFP to GFP FRET wide excitation (31034), GFP wide blue excitation (31054)
565	Qdot® 565 filter set (20 nm EM; 32005) (460SPUV/475DCXRU/D565/20nm) Qdot® 565 filter set (40 nm EM; 32009) (460SPUV/475DCXRU/D565/40nm)	Eosin (41011), Cascade Yellow™ (31038), JP2(YGFP with EGFP-31040, Auramine (31015)
585	Qdot® 585 filter set (20 nm EM; 32004) (460SPUV/475DCXRU/D585/20nm) Qdot® 585 filter set (40 nm EM; 32008) (460SPUV/475DCXRU/D585/40nm)	R-PE (41003), Rhodamine LP (41032, FITC/PI (41016)
605	Qdot® 605 filter set (20 nm EM; 32003) (460SPUV/475DCXRU/D605/20nm) Qdot® 605 filter set (40 nm EM; 32007) (460SPUV/475DCXRU/D605/40nm)	Cy3 narrow excitation (41007a), Texas Red®/Cy3.5 (31004), TRITC (41002, 41002a, 41002b), Ethidium Bromide (41006)
655	Qdot® 655 filter set (20 nm EM; 32011) (460SPUV/475DCXRU/D655/20nm) Qdot® 655 filter set (40 nm EM; 32012) (460SPUV/475DCXRU/D655/40nm)	Texas Red® (41004), Propidium iodide (41005), Fura Red™ (31012), Chlorophyll (31017), Allophycocyanin (31006)
705*	Qdot® 705 filter set (20 nm EM; 32014) (460SPUV/475DCXRU/D705/20nm) Qdot® 705 filter set (40 nm EM; 32015) (460SPUV/475DCXRU/D705/40nm)	Cy5 Longpass (41024), Cy5 (41008), Cy5 narrow excitation (41033), Cy5.5 (41023), Alexa Fluor® 680 (41042), Cy5.5 (red-shifted; 41022)
800*	Qdot® 800 filter set (30 nm EM; 32020) (460SPUV/475DCXRU/D800/30nm) Qdot® 800 filter set (50 nm EM; 32021) (460SPUV/475DCXRU/D800/50nm)	Cy7 (41009), Li-Cor for IRDye 800 (41037), Cy7 (SP106)
All colorst	Qdot® Multiple Emission Set (71014) (460SPUV, 475DCXRU, D525/20nm, D605/20nm, D565/20nm, D585/20nm)	UV (11000V2), Blue/Violet (11003V2), UV/Violet (11011V2)

*The 705 and 800 nm quantum dot emission cannot be seen by eye and must be detected with an IR-sensitive detector.

†For viewing multiple colors of Qdot® nanocrystals through microscope eyepieces.

Appendix 3: Extinction Coefficients

Table 5. Extinction coefficients of Qdot® streptavidin conjugates at common excitation wavelengths.

Product	350 nm, in $\text{cm}^{-1}\text{M}^{-1}$	405 nm, in $\text{cm}^{-1}\text{M}^{-1}$	488 nm, in $\text{cm}^{-1}\text{M}^{-1}$	532 nm, in $\text{cm}^{-1}\text{M}^{-1}$
Qdot® 525 nanocrystals	710,000	360,000	130,000	Not applicable
Qdot® 565 nanocrystals	1,900,000	1,100,000	290,000	139,000
Qdot® 585 nanocrystals	3,500,000	2,200,000	530,000	305,000
Qdot® 605 nanocrystals	4,400,000	2,800,000	1,100,000	580,000
Qdot® 625 nanocrystals	14,700,000	9,900,000	2,700,000	870,000
Qdot® 655 nanocrystals	9,100,000	5,700,000	2,900,000	2,100,000
Qdot® 705 nanocrystals	12,900,000	8,300,000	3,000,000	2,100,000
Qdot® 800 nanocrystals	12,600,000	8,000,000	3,000,000	2,000,000

Appendix 4: Bibliography

There are a number of references that describe the size-dependent properties of the semiconductor nanocrystals. These range in complexity from fairly straightforward descriptions to fairly comprehensive mathematical and physical descriptions of the optical properties. In addition, we have included some representative references that describe the core-shell structures, and the improved chemical properties that are obtained through such structures. References 8–11 describe quantum dots and FRET:

1. *Sci Am* 285, 66 (2001); 2. *J Phys Chem B* 100, 13226 (1996); 3. *J Am Chem Soc* 115, 8706 (1993); 4. *Phys Rev B* 53, 16338 (1996); 5. *J Phys Chem* 100, 468 (1996); 6. *J Phys Chem B* 101, 9463 (1997); 7. *J Am Chem Soc* 119, 7019 (1997); 8. *Nano Lett* 1, 469 (2001); 9. *J. Am. Chem. Soc* 126, 301 (2004); 10. *Nat Mater* 2, 630 (2003); 11. *Nat Biotechnol* 21, 1387 (2003).

A number of references describe the biological properties of some quantum dots used in experiments. These papers are selected to represent some of the different classes of applications, but this list is not exhaustive. These materials are all quite different from the Qdot® conjugates that are sold by Invitrogen, and the results are not necessarily representative of results attainable with these materials:

12. *Science* 281, 2013 (1998); 13. *Science* 281, 2016 (1998); 14. *J Am Chem Soc* 124, 4586 (2002); 15. *Proc Natl Acad Sci U S A*. 99, 12617 (2002); 16. *Science* 298, 1759 (2002); 17. *Nat Biotechnol* 21, 41 (2003); 18. *Nat Biotechnol* 21, 47 (2003).

Also of interest:

19. Hermanson, GT. *Bioconjugate Techniques*, Academic Press, 1996; 20. Lakowicz, J. *Principles of Fluorescence Spectroscopy*. Kluwer Academic Publishing, 1999; 21. *J Am Chem Soc* 128, 12800 (2006); 22. Colligan et. al. *Current Protocols in Immunology*. J. Wiley, Annually Updated, 2002.

Product List Current prices may be obtained from our website or from our Customer Service Department.

Cat no.	Product Name	Unit Size
A10196	Qdot® 625 streptavidin conjugate *1 µM solution*	200 µL
Q10101MP	Qdot® 605 streptavidin conjugate *1 µM solution*	200 µL
Q10103MP	Qdot® 605 streptavidin conjugate, starter kit *1 µM solution*	50 µL
Q10111MP	Qdot® 585 streptavidin conjugate *1 µM solution*	200 µL
Q10113MP	Qdot® 585 streptavidin conjugate, starter kit *1 µM solution*	50 µL
Q10121MP	Qdot® 655 streptavidin conjugate *1 µM solution*	200 µL
Q10123MP	Qdot® 655 streptavidin conjugate, starter kit *1 µM solution*	50 µL
Q10131MP	Qdot® 565 streptavidin conjugate *1 µM solution*	200 µL
Q10133MP	Qdot® 565 streptavidin conjugate, starter kit *1 µM solution*	50 µL
Q10141MP	Qdot® 525 streptavidin conjugate *1 µM solution*	200 µL
Q10143MP	Qdot® 525 streptavidin conjugate, starter kit *1 µM solution*	50 µL
Q10151MP	Qdot® Streptavidin Sampler Kit *1 µM solutions*	1 kit
Q10161MP	Qdot® 705 streptavidin conjugate *1 µM solution*	200 µL
Q10163MP	Qdot® 705 streptavidin conjugate, starter kit *1 µM solution*	50 µL
Q10171MP	Qdot® 800 streptavidin conjugate *1 µM solution*	200 µL
Q10173MP	Qdot® 800 streptavidin conjugate, starter kit *1 µM solution*	50 µL
A1310	7-aminoactinomycin D (7-AAD)	1 mg
A21355	anti-OxPhos Complex V inhibitor protein, mouse IgG1, monoclonal 5E2 (anti-ATP synthase IP; anti-F1F0-ATPase IP) *human mitochondrial reactivity*	100 µg
E21390	Endogenous Biotin-Blocking Kit *100 assays*	1 kit
P3356	Propidium iodide *1.0 mg/mL solution in water*	10 mL
S33025	SelectFX® Nuclear Labeling Kit *DAPI, SYTOX® Green, 7-AAD, TO-PRO®-3 iodide* *for fixed cells*	1 kit
S7020	SYTOX® Green nucleic acid stain *5 mM solution in DMSO*	250 µL
T3605	TO-PRO®-3 iodide (642/661) *1 mM solution in DMSO*	1 mL
39-6400	Mouse anti-Nucleolin	100 µg
70011-044	Phosphate-Buffered Saline (PBS), pH 7.4 (10X), liquid	500 mL

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Qdot® ITK™ Carboxyl Quantum Dots

Catalog nos. Q21341MP, Q21391MP, Q21331MP, Q21311MP, Q21301MP, A10200, Q21321MP, Q21361MP, Q21371MP

Table 1. Contents and storage information.

Material	Amount	Concentration	Storage	Stability
Qdot® ITK™ carboxyl quantum dots	250 µL	8 µM solution in 50 mM borate, pH 9.0	<ul style="list-style-type: none"> • 2–6°C • Do not freeze 	When stored as directed the product is stable for at least 6 months.
Approximate fluorescence excitation/emission spectra: See Figure 2.				

Introduction

Structure of Qdot® Nanocrystals

Qdot® ITK™ (Innovator's Tool Kit) carboxyl quantum dots are made from nanometer-scale crystals of a semiconductor material (CdSe), which are shelled with an additional semiconductor layer (ZnS) to improve their chemical and optical properties. The Qdot® 705 and Qdot® 800 nanocrystals, which include CdSeTe, are made in a similar fashion. These materials have narrow, symmetric emission bands with emission maxima near 525 nm, 565 nm, 585 nm, 605 nm, 625 nm, 655 nm, 705 nm, or 800 nm. This core-shell material (Figure 1A) is further coated with a polymer layer that allows facile dispersion of the quantum dots in aqueous solutions with retention of their optical properties. The polymer coating has –COO⁻ surface groups available for modifications such as macromolecule attachment (Figure 1B). Qdot® ITK™ carboxyl quantum dots are about the size of a large macromolecule or protein.

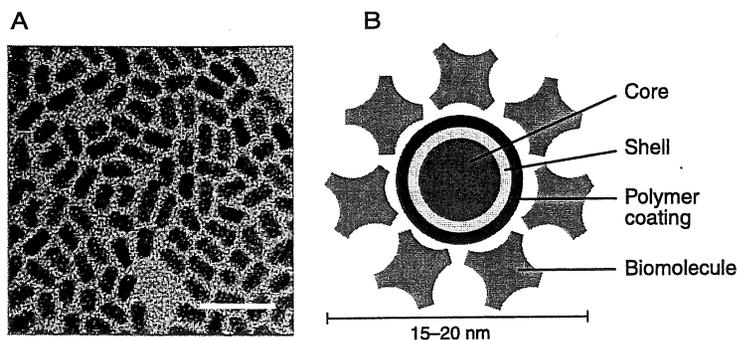


Figure 1. A. Transmission electron microscope image of core-shell Qdot® nanoparticles at 200,000x magnification. Scale bar = 20 nm. B. Schematic of the overall structure of a Qdot® conjugate. The layers represent the distinct structural elements of the Qdot® nanocrystal conjugates, and are roughly to scale.

Optical Properties

The optical properties of Qdot® nanocrystals are different from those of typical organic dye molecules. The colors of light that Qdot® nanocrystals emit are strongly dependent on particle size, creating a common platform of fluorescent labels emitting from green to the near IR, all manufactured from the same underlying semiconductor material (see *Bibliography*, references 1–11 in the *Appendix*). The size of Qdot® nanocrystals is tightly controlled in the production process, resulting in materials with narrow and symmetric emission bands and that are extremely bright and photostable. While the fluorescence emission bands from the Qdot® 705 and Qdot® 800 nanocrystals are broader than the emission bands of the visible wavelength Qdot® nanocrystals, all the Qdot® fluorophores have strong emission intensity and superior photostability. Note that Qdot® 705 and 800 nm emissions cannot be seen by eye, but are easily detected by many cameras and detectors. These properties are exploited in a variety of immunofluorescence techniques, and can result in substantially better results than are attainable with conventional fluorescent labels (see *Bibliography*, references 12–18). Though these materials are compatible with a number of standard fluorescence techniques, there are some novel aspects of their chemistry and detection that require careful consideration to obtain optimal results.

Spectral Characteristics

Organic fluorescent dyes have excitation and emission spectra with a relatively small Stokes shift, which means that the optimal excitation wavelength is close to the emission peak. Filter sets used with fluorescent dyes reflect this characteristic.¹⁹ Light absorption efficiency of Qdot® nanocrystals increases dramatically to the blue of the emission (Figure 2). These unique spectral properties are due to the semiconductor material that makes up the core of the Qdot® nanocrystals, which gives rise to both their absorption and emission properties. (see *Bibliography*, references 1–11). Despite their broad wavelength range of light absorption, the emission wavelength of these materials is independent of the excitation wavelength. For example, whether exciting at 400 nm or 633 nm, the shape of the emission band of Qdot® 655 nanocrystals remains the same, while the intensity is approximately 11-fold higher with 400 nm excitation. Light absorption and consequent excitation at shorter wavelength, with fixed emission, results in a large “apparent Stokes shift.” Short wavelength excitation improves sensitivity by reducing autofluorescence and takes advantage of the inherently greater light absorption of these materials in the blue to violet spectral region, greatly simplifying simultaneous, multiplexed detection of several Qdot® nanocrystal colors. See *Appendix 3* for extinction coefficients of the different materials at common excitation wavelengths.

Optical Filter Selection

To achieve the optimal signal from Qdot® conjugates, we recommend using Qdot® optimized filter sets that are available from Omega Optical, Semrock, or Chroma Technology Corporation (see *Appendix 2* for details).

Qdot® conjugates can also be viewed through some standard filter sets, albeit with lower detection efficiency and reduced brightness. For example, three Omega Optical standard filter sets capable of detecting Qdot® 705 conjugates are XF140-2 (Alexa Fluor® 633 & Alexa Fluor® 647), XF70 (Alexa Fluor® 660 & Cy5), and XF141-2 (Cy5.5). Visualization of Qdot® conjugates using a custom filter set is preferred because excitation and detection is less efficient using filters that have not been selected specifically for use with Qdot® conjugates. Using a custom filter set, Qdot® 605 conjugate signal is approximately five times as bright as it is using the TRITC filter set, and approximately ten times brighter than it is using the Texas Red®/Cy3.5 filter set (Figure 3). Qdot® optimal filters and standard filter sets are available from different filter manufacturers. *Appendix 2* illustrates some common filter sets and the optimal filter set recommendations for the available Qdot® conjugates. Use of the optimal filter set is critical for attaining optimal signal and sensitivity in your experiments.

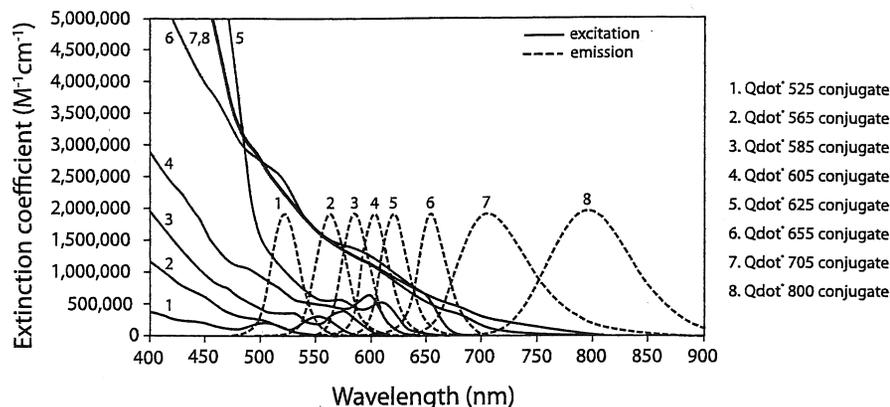


Figure 2. Typical absorption and emission spectra of Qdot® 525 conjugate (1), Qdot® 565 conjugate (2), Qdot® 585 conjugate (3), Qdot® 605 conjugate (4), Qdot® 625 conjugate (5), Qdot® 655 conjugate (6), Qdot® 705 conjugate (7), Qdot® 800 conjugate (8).

Before You Begin

Qdot® nanocrystals have chemical and optical properties that provide significant advantages over conventional fluorophores in both sensitivity and stability in fluorescent labeling and tracking applications. Qdot® ITK™ carboxyl quantum dots are used in a wide variety of labeling and tracking applications, including preparation and use of peptide derivatives,²⁰ nucleic acid conjugates,^{21,22} polysaccharide conjugates,²³ in stem cell tracking²⁴ and for other uses in which ultrabright and stable fluorescence is desired.

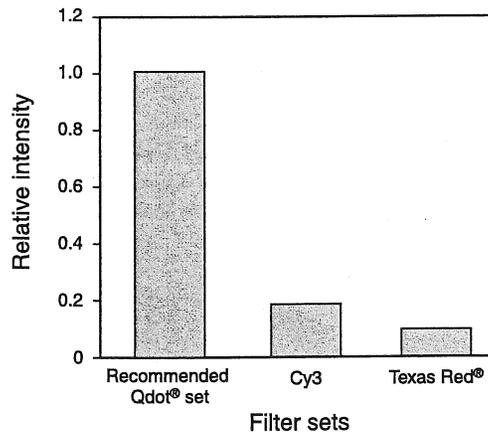


Figure 3. Detection of Qdot® conjugates on tissue sections with recommended and standard filter sets. Mouse kidney sections were stained with Qdot® 605 conjugate, and then images were collected on a Nikon epi-fluorescence microscope in 16 bit capture mode. The mean fluorescence of positively stained samples was extracted using Scion Image software. The recommended Qdot® filter set included a 460 nm short pass exciter, a 475 nm dichroic, and a 605/20 nm band pass emitter. The Cy3 filter set included a 545/30 nm exciter, a 570 nm dichroic, and a 610/75 nm emitter. The Texas Red® filter set included a 560/40 nm exciter, a 595 nm dichroic, and a 630/60 nm emitter.

For additional applications such as immunocytochemistry, tissue section staining, western blotting, as well as multiplexing using Qdot® streptavidin and secondary antibody conjugates, download the Qdot® Conjugates Protocol Handbook from www.invitrogen.com. For additional information and useful protocols for various applications with Qdot® nanocrystals and their conjugates, see *Quantum Dots: Applications in Biology (Methods in Molecular Biology)*.²⁵

Note: The near infrared 705 and 800 nm quantum dot emissions cannot be seen by eye, but are easily detected by many cameras and detectors.

General Considerations

Buffer compatibility

In our experience, Qdot® ITK™ carboxyl quantum dots and many of their conjugates have stable emission in a number of buffers, and the quantum yield and colloidal dispersion of conjugates made with these materials has been found to be stable at physiological and near-physiological pH (not investigated outside this range) in Tris, HEPES, phosphate, and borate buffers. In addition, a number of surfactants and additives such as Tween 20, Triton® X-100, and EDTA, among others, have been shown to maintain nanocrystal fluorescence when used at up to 0.5% concentration. We recommend storage of the Qdot® ITK™ carboxyl quantum dot product at the concentration at which it is shipped, rather than at high dilution. Storage of Qdot® ITK™ carboxyl quantum dots and their macromolecule conjugates at working dilution may result in substantial performance degradation. While we have not characterized the stability of all Qdot® ITK™ carboxyl quantum dots under all of these conditions, we anticipate similar levels of stability across the range of product colors.

Qdot® nanocrystal toxicity

We have not investigated the toxicity of Qdot® ITK™ carboxyl quantum dots. The materials are provided in a solution which is ~2 mM total Cd concentration; however, the CdSe core is encapsulated in a crystalline shell of ZnS and the amphiphilic polymer coating, which may help prevent formation of free Cd. We have demonstrated the utility of these materials in a variety of live-cell *in vitro* labeling experiments, but do not have systematic data on the toxicity of the materials to humans, to animals, or to cells in culture.

FRET or close-proximity quenching

We have not systematically investigated the energy transfer properties of the Qdot® nanocrystals, though they may have useful properties as energy transfer donors and acceptors. We have investigated the fluorescence of Qdot® 605 conjugates which are coupled to each other through a bis-biotin linker, and found that the emission intensity of the materials was unperturbed at any concentration of biotin cross-linker. These results suggest that the interparticle quenching of these Qdot® conjugates is negligible. Published literature indicates that Qdot® nanocrystals can be used as energy acceptors in time-resolved FRET (TR-FRET) studies.²⁶

Disposal of Qdot® Conjugate

The Qdot® conjugate contains cadmium and selenium in an inorganic crystalline form. Dispose of the material in compliance with all applicable local, state, and federal regulations for disposal of these classes of material. For more information on the composition of these materials, consult the Material Safety Data Sheet.

Conjugating Qdot® ITK™ carboxyl quantum dots

A protocol for conjugating streptavidin to Qdot® ITK™ carboxyl quantum dots is supplied below. You can use similar methods to conjugate other proteins and biomolecules of interest, providing they are compatible with the coupling chemistry described below or with alternate conjugation chemistry under consideration. The amounts suggested below may need to be adjusted to accommodate your conjugation requirements.

Conjugation of Qdot® ITK™ Carboxyl Quantum Dots to Streptavidin

Materials Required but Not Provided

- 10 mg N-ethyl-N'-dimethylaminopropyl-carbodiimide (EDC)
- 10 mg/mL Streptavidin (Invitrogen Cat. no. S888) in 10 mM borate buffer, pH 7.4
- 10 mM borate buffer, pH 7.4
- 50 mM borate buffer, pH 8.3
- Ultrafiltration units with 100 kDa cutoff, size 4 mL (Amicon Ultra-4—Millipore Cat. no. UFC810008) or size 15 mL (Amicon Ultra 15—Millipore Cat. no. UFC910008)
- Filter syringes: Acrodisc® 25 mm PF Syringe Filter with 0.8/0.2 µm Supor® Membrane or Acrodisc® Syringe Filter 0.2 µm Supor® Membrane Low Protein Binding Non Pyrogenic
- PES (polyethersulfone) syringe filters, 0.2 µm (Whatman Cat. no. 6876-2502)

Preparing Streptavidin Solution

Prepare a 10 mg/mL streptavidin solution in 10 mM borate buffer, pH 7.4. Mix well. You will need 80 nmol of streptavidin for the conjugation reaction.

Amount

The molar concentration of each reagent used during the conjugation protocol is described below. Use these concentrations for the initial experiments and based on your results, you may need to optimize these concentrations to obtain the desired level of conjugation.

Reagent	Concentration	Equivalent
Qdot® Reagent	2 nmol (250 µL at 8 µM concentration)	1
Streptavidin	80 nmol (0.12–4.8 mg at 10 mg/mL)	40
EDC	3000 nmol (0.57 mg at 10 mg/mL)	1500

Experimental Protocol

Conjugation Protocol

Please read the entire protocol before starting.

- 1.1 In a small glass vial with a small stirbar, dilute 250 µL of 8 µM stock solution of Qdot® ITK™ carboxyl quantum dots to 2 mL using 10 mM borate buffer, pH 7.4. Mix well by stirring.
- 1.2 Add 0.48 mL of 10 mg/mL streptavidin to the Qdot® ITK™ carboxyl quantum dots reagent (step 1.1). Continue stirring.
- 1.3 Weigh ~5 mg of EDC in a 1.5 mL microcentrifuge tube and add 0.5 mL deionized water to obtain a 10 mg/mL EDC stock solution. Prepare EDC solution just before use.
- 1.4 Immediately, add 57 µL of 10 mg/mL EDC stock solution to the Qdot® solution (step 1.2).
- 1.5 Stir gently for 1–2 hours at room temperature for the conjugation.
- 1.6 Filter the conjugate solution through a 0.2 µm PES syringe filter to remove any large aggregates and transfer the solution to a clean centrifugal ultrafiltration unit (100 kDa cutoff).
- 1.7 Centrifuge at the recommended speed for the ultrafiltration unit for at least 5 buffer exchanges using 50 mM borate buffer, pH 8.3 to remove any excess unbound protein. Ensure

that the volume of concentration is >10-fold (e.g., 4 mL to <400 μ L) each time.

- 1.8 After ultracentrifugation is complete, filter the solution through a 0.2 μ m syringe filter or a 0.8/0.2 μ m combination syringe filter to remove any aggregates. Store the Qdot[®] conjugate solution at 4°C. **Do not freeze** the nanocrystal conjugate.

Appendix 1: Troubleshooting Guide

The properties of Qdot[®] conjugates are different from fluorescent dyes and may require slight modifications to current protocols. We've included this section to help with some specific issues that may arise while using these materials.

No Signal

Optical Setup suitability

Make sure that you are using an appropriate filter set to detect the signals. See *Appendix 2* for a list of appropriate and optimal filters for the Qdot[®] conjugates. Contact Technical Support (probetech@invitrogen.com) for more details on particular filter set requirements.

Qdot[®] conjugate luminosity

Qdot[®] conjugates normally fluoresce brightly under a hand-held ultraviolet lamp (long wave, such as the type used to visualize ethidium bromide on agarose gels). The 705 and 800 nm quantum dot emission cannot be seen by eye, but is detected by many cameras and detectors. Though we have not seen pronounced loss of fluorescence of these materials under any storage conditions that we have investigated, we have not been able to examine all storage conditions. If the Qdot[®] conjugates do not appear to fluoresce under the long wave UV excitation, contact Technical Support (probetech@invitrogen.com) for assistance.

Appendix 2: Optimal Usable Filter Sets for Qdot® Conjugates

Table 3. Omega optical filter set for Qdot® conjugates.

Color	Optimal filter sets	Usable filter sets
525	XF301 Qdot®525 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP Emitter: 525WB20)	XF100-3, XF100-2, XF115-2, XF89-2
565	XF302 Qdot® 565 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter 565WB20)	XF104-2, XF105-2
585	XF303 Qdot® 585 filter set (Exciter: 1 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 585WB20)	XF101-2, XF137-2, XF152-2
605	XF304 Qdot® 605 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 605WB20)	XF108-2, XF102-2, XF103-2
655	XF305 Qdot® 655 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 655WB20)	XF102-2, XF40-2, XF42, XF45
705*	XF306 Qdot® 705 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 710AF40)	XF140-2, XF70, XF110-2, XF141-2, XF48-2
800 *	XF307 Qdot® 800 filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 800WB80)	XF308 Qdot® 800 filter set for multiplexing (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP/ Emitter: 840WB80)
All colorst	XF300 Qdot® filter set (Exciter 1: 425DF45 or Exciter 2: 415WB100/ Dichroic: 475DCLP Emitters: 800WB80, 840WB80, 710AF40, 655WB20, 605WB20, 585WB20, 565WB20, and 525WB20)	XF129-2, XF130-2

*The 705 and 800 nm quantum dot emission cannot be seen by eye and must be detected with an IR-sensitive detector.

†For viewing multiple colors of Qdot® nanocrystals through microscope eyepieces.

Table 4. Semrock filter sets for Qdot® conjugates.

Color	Optimal filter sets	Usable filter sets
525	BrightLine® QD525-A Filter Sets: QD525-A-000 or QD525-A-000-ZERO (Exciter: FF01-435/40-25) (Dichroic: FF510-Di01-25 x 36) (Emitter: FF01-525/15-25)	GFP-3035B
565	--	FITC-3504B or YFP-2427A
585	--	TRITC-A
605	BrightLine® QD605-A Filter Sets: QD605-A-000 or QD605-A-000-ZERO (Exciter: FF01-435/40-25) (Dichroic: FF510-Di01-25 x 36) (Emitter: FF01-605/15-25)	TRITC-A
625	BrightLine® QD625-A Filter Sets: QD625-A-000 or QD625-A-000-ZERO (Exciter: FF01-435/40-25) (Dichroic: FF510-Di01-25 x 36) (Emitter: FF01-625/15-25)	Texas Red® (4040B)
655	BrightLine® QD655-A Filter Sets: QD655-A-000 or QD655-A-000-ZERO (Exciter: FF01-435/40-25) (Dichroic: FF510-Di01-25 x 36) (Emitter: FF01-655/15-25)	Texas Red® (4040B)
705*	--	Cy5-4040A or Cy5.5-A
800*	--	Cy7-A
LP multi†	QDLP-A Filter Set: QDLP-A-000 (Exciter: FF01-435/40-25) (Dichroic: FF510-Di01-25 x 36) (Emitter: FF01-500/LP-25)	CFW-LP01-CLINICAL

*The 705 and 800 nm quantum dot emission cannot be seen by eye and must be detected with an IR-sensitive detector.
†For viewing multiple colors of Qdot® nanocrystals through microscope eyepieces.

Table 5. Chroma Technology filter sets for Qdot® conjugates.

Color	Optimal filter sets	Usable filter sets
525	Qdot® 525 filter set (20 nm EM; 32006) (460SPUV/475DCXRU/D525/20nm) Qdot® 525 filter set (40 nm EM; 32010) (460SPUV/475DCXRU/D525/40nm)	FITC/RSGFP/Bodipy®/Fluo-3/DiO (41001), FITC/RSGFP Longpass (40012), BFP to GFP FRET (31032), BFP to GFP FRET wide excitation (31034), GFP wide blue excitation (31054)
565	Qdot® 565 filter set (20 nm EM; 32005) (460SPUV/475DCXRU/D565/20nm) Qdot® 565 filter set (40 nm EM; 32009) (460SPUV/475DCXRU/D565/40nm)	Eosin (41011), Cascade Yellow™ (31038), JP2(YGFP with EGFP-31040, Auramine (31015)
585	Qdot® 585 filter set (20 nm EM; 32004) (460SPUV/475DCXRU/D585/20nm) Qdot® 585 filter set (40 nm EM; 32008) (460SPUV/475DCXRU/D585/40nm)	R-PE (41003), Rhodamine LP (41032, FITC/PI (41016)
605	Qdot® 605 filter set (20 nm EM; 32003) (460SPUV/475DCXRU/D605/20nm) Qdot® 605 filter set (40 nm EM; 32007) (460SPUV/475DCXRU/D605/40nm)	Cy3 narrow excitation (41007a), Texas Red®/Cy3.5 (31004), TRITC (41002, 41002a, 41002b), Ethidium Bromide (41006)
655	Qdot® 655 filter set (20 nm EM; 32011) (460SPUV/475DCXRU/D655/20nm) Qdot® 655 filter set (40 nm EM; 32012) (460SPUV/475DCXRU/D655/40nm)	Texas Red® (41004), Propidium iodide (41005), Fura Red™ (31012), Chlorophyll (31017), Allophycocyanin (31006)
705*	Qdot® 705 filter set (20 nm EM; 32014) (460SPUV/475DCXRU/D705/20nm) Qdot® 705 filter set (40 nm EM; 32015) (460SPUV/475DCXRU/D705/40nm)	Cy5 Longpass (41024), Cy5 (41008), Cy5 narrow excitation (41033), Cy5.5 (41023), Alexa Fluor® 680 (41042), Cy5.5 (red-shifted; 41022)
800*	Qdot® 800 filter set (30 nm EM; 32020) (460SPUV/475DCXRU/D800/30nm) Qdot® 800 filter set (50 nm EM; 32021) (460SPUV/475DCXRU/D800/50nm)	Cy7 (41009), Li-Cor for IRDye 800 (41037), Cy7 (SP106)
All colorst	Qdot® Multiple Emission Set (71014) (460SPUV, 475DCXRU, D525/20nm, D605/20nm, D565/20nm, D585/20nm)	UV (11000V2), Blue/Violet (11003V2), UV/Violet (11011V2)

*The 705 and 800 nm quantum dot emission cannot be seen by eye and must be detected with an IR-sensitive detector.

†For viewing multiple colors of Qdot® nanocrystals through microscope eyepieces.

Appendix 3: Extinction Coefficients

Table 6. Extinction coefficients of Qdot® conjugates at common excitation wavelengths.

Product	350 nm, in $\text{cm}^{-1}\text{M}^{-1}$	405 nm, in $\text{cm}^{-1}\text{M}^{-1}$	488 nm, in $\text{cm}^{-1}\text{M}^{-1}$	532 nm, in $\text{cm}^{-1}\text{M}^{-1}$
Qdot® 525 nanocrystals	710,000	360,000	130,000	Not applicable
Qdot® 565 nanocrystals	1,900,000	1,100,000	290,000	139,000
Qdot® 585 nanocrystals	3,500,000	2,200,000	530,000	305,000
Qdot® 605 nanocrystals	4,400,000	2,800,000	1,100,000	580,000
Qdot® 625 nanocrystals	14,700,000	9,900,000	2,700,000	870,000
Qdot® 655 nanocrystals	9,100,000	5,700,000	2,900,000	2,100,000
Qdot® 705 nanocrystals	12,900,000	8,300,000	3,000,000	2,100,000
Qdot® 800 nanocrystals	12,600,000	8,000,000	3,000,000	2,000,000

Appendix 4: Bibliography

There are a number of references that describe the size-dependent properties of the semiconductor nanocrystals. These range in complexity from fairly straightforward descriptions to fairly comprehensive mathematical and physical descriptions of the optical properties. In addition, we have included some representative references that describe the core-shell structures, and the improved chemical properties that are obtained through such structures. References 8–11 describe quantum dots and FRET:

1. *Sci Am* 285, 66 (2001); 2. *J Phys Chem B* 100, 13226 (1996); 3. *J Am Chem Soc* 115, 8706 (1993); 4. *Phys Rev B* 53, 16338 (1996); 5. *J Phys Chem* 100, 468 (1996); 6. *J Phys Chem B* 101, 9463 (1997); 7. *J Am Chem Soc* 119, 7019 (1997); 8. *Nano Lett* 1, 469 (2001); 9. *J. Am. Chem. Soc* 126, 301 (2004); 10. *Nat Mater* 2, 630 (2003); 11. *Nat Biotechnol* 21, 1387 (2003).

A number of references describe the biological properties of some quantum dots used in experiments. These papers are selected to represent some of the different classes of applications, but this list is not exhaustive. These materials are all quite different from the Qdot® conjugates that are sold by Invitrogen, and the results are not necessarily representative of results attainable with these materials:

12. *Science* 281, 2013 (1998); 13. *Science* 281, 2016 (1998); 14. *J Am Chem Soc* 124, 4586 (2002); 15. *Proc Natl Acad Sci U S A*. 99, 12617 (2002); 16. *Science* 298, 1759 (2002); 17. *Nat Biotechnol* 21, 41 (2003); 18. *Nat Biotechnol* 21, 47 (2003).

Also of interest:

19. Lakowicz, J. *Principles of Fluorescence Spectroscopy*. Kluwer Academic Publishing, 1999; 20. *Conf Proc IEEE Eng Med Biol Soc* 1, 1470 (2006); 21. *J Fluoresc* 17, 193 (2007); 22. *Mol Cell Probes* 21, 116 (2006); 23. Kim J, Park, K and Hahn S K, *Int J Biol Macromol*, in press (2007); 24. *Stem Cells* 25, 2128 (2007); 25. Hotz, CZ., and Bruchez, M. *Quantum Dots: Applications in Biology (Methods in Molecular Biology)* 2007. 26. *J Am Chem Soc* 128, 12800 (2006).

Product List Current prices may be obtained from our website or from our Customer Service Department.

Cat no.	Product Name	Unit Size
Q21341MP	Qdot® 525 ITK™ carboxyl quantum dots *8 µM solution*	250 µL
Q21391MP	Qdot® 545 ITK™ carboxyl quantum dots *8 µM solution*	250 µL
Q21331MP	Qdot® 565 ITK™ carboxyl quantum dots *8 µM solution*	250 µL
Q21311MP	Qdot® 585 ITK™ carboxyl quantum dots *8 µM solution*	250 µL
Q21301MP	Qdot® 605 ITK™ carboxyl quantum dots *8 µM solution*	250 µL
A10200	Qdot® 625 ITK™ carboxyl quantum dots *8 µM solution*	250 µL
Q21321MP	Qdot® 655 ITK™ carboxyl quantum dots *8 µM solution*	250 µL
Q21361MP	Qdot® 705 ITK™ carboxyl quantum dots *8 µM solution*	250 µL
Q21371MP	Qdot® 800 ITK™ carboxyl quantum dots *8 µM solution*	250 µL
Related products		
S888	Streptavidin.....	5 mg

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Qtracker® Cell Labeling Kits

Catalog nos. A10198, Q25001MP, Q25011MP, Q25021MP, Q25031MP, Q25041MP, Q25061MP, Q25071MP

Table 1. Contents and storage information.

Material	Amount	Concentration	Storage	Stability
Qtracker® nanocrystals, Component A	100 µL	2 µM in 50 mM borate buffer, pH 8.3	<ul style="list-style-type: none"> • 2–6°C • DO NOT FREEZE 	When stored as directed the product is stable for at least 6 months.
Qtracker® carrier, Component B	100 µL	Phosphate buffered saline (PBS), pH 7.2		

Approximate fluorescence excitation and emission maxima: See Table 2.

Introduction

Qtracker® Cell Labeling Kits are designed for labeling cells grown in culture with highly fluorescent Qdot® nanocrystals. Once inside the cells, Qtracker® labels provide intense, stable fluorescence that can be traced through several generations, and are not transferred to adjacent cells in a population.

Qtracker® Cell Labeling Kits are available in seven colors—525 nm, 565 nm, 585 nm, 605 nm, 625 nm, 655 nm, 705 nm, or 800 nm emission—and are excellent tools for long-term tracking or imaging studies of live cells, including migration, motility, morphology, and other cell function assays.

The Qtracker® Cell Labeling Kits use a custom targeting peptide^{1,2} to deliver Qdot® nanocrystals into the cytoplasm of live cells. Cytoplasmic delivery by this mechanism is not mediated by a specific enzyme; therefore, no cell-type specificity has been observed. Delivery is typically accomplished in less than 1 hour. Qdot® nanocrystals delivered by the Qtracker® Cell Labeling Kits are compatible with serum-sensitive cells; intense fluorescence is maintained in complex cellular environments and under various biological conditions including changes in intracellular pH, temperature, and metabolic activity. Furthermore, autofluorescence commonly observed in cells or tissues can be avoided using Qtracker® 655, 705, or 800 Kits.

Features and Applications

Using Qtracker® Cell Labeling Kits, you can observe labeled cells using extensive continuous illumination, without the photobleaching and degradation problems often associated with organic dyes.^{3–5} Qtracker® labels are distributed in vesicles in the cytoplasm (Figure 1), and are inherited by daughter cells for at least six generations. Fluorescence from the Qtracker® labels can be seen up to a week after delivery in some cell lines. Long-term cellular retention makes Qtracker® Cell Labeling Kits ideal for studying cell motility (Figure 2), migration,

differentiation, morphology, and many other cellular function studies.^{3,4} Qtracker® labels do not leak out of intact cells to be taken up by adjacent cells in the population (Figure 3).

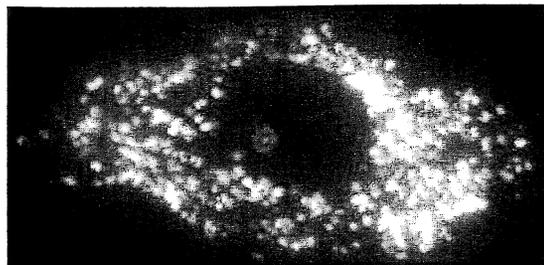


Figure 1. Distribution of Qtracker® labels in the cytoplasm: HeLa cells labeled with the Qtracker® 655 Cell Labeling Kit were observed with a Leica TCS SP2 confocal microscopy to see the distribution of the Qtracker® reagent in the cytoplasm (excitation at 488 nm).

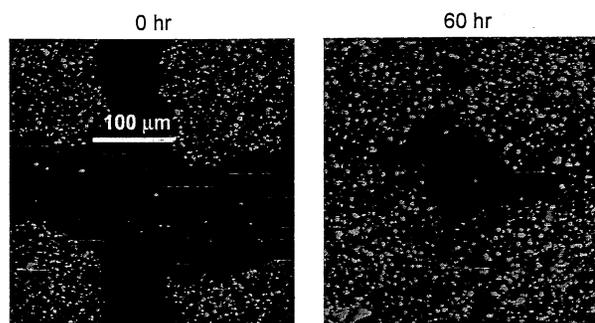


Figure 2. Motility of HeLa cells: A monolayer of HeLa cells was labeled with the Qtracker® 655 Cell Labeling Kit. The gap was made by scratching with a 200 μ L pipette tip (\sim 1 mm) and imaged using a Leica TCS SP2 confocal microscope (excitation at 488 nm; 10X objective). The cells moved to fill the gap and retained normal motility.

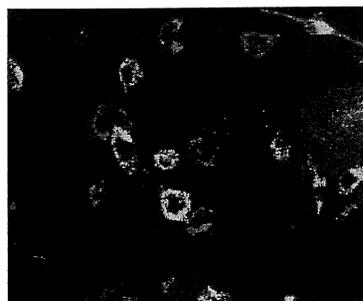


Figure 3. U-118 and HeLa cells were labeled with Qtracker® 565 and 655 Cell Labeling Kits, respectively, and co-cultured for 24 hours. The image was captured using a Leica TCS SP2 confocal microscope. Both colors are easily resolved, well retained, and well segregated in their respective cell lines.

Imaging Platforms

Qtracker® reagent-labeled live cells can be easily monitored on a variety of platforms, including flow cytometry, fluorescence/confocal microscopy, fluorescence microplate readers, and high-content imaging systems.

Cytotoxicity and Viability Studies

The cytotoxicity of the materials use in Qtracker® Cell Labeling Kits has been tested in a variety of cell lines including CHO, HeLa, U-118, 3T3, HUVEC, and Jurkat cells. Labeling with Qtracker® Cell Labeling Kits appears to exert minimal impact on cellular surface marker expression, cell proliferation, cellular enzyme activity, and cell motility; no effect on the CD3 expression level of Jurkat cells

was observed following labeling with the Qtracker® Cell Labeling Kit.

The effect of Qdot® nanocrystal loading on cellular viability has been examined using cell proliferation and cellular enzyme activity measurements (Figure 4). The results indicate that labeling with the Qtracker® Cell Labeling Kit has no significant effect on cell proliferation and cellular enzyme activity.

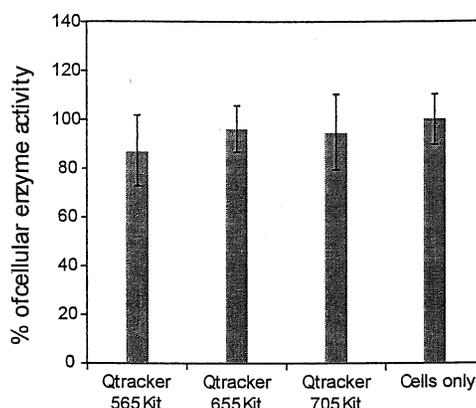


Figure 4. Effect of Qtracker® labeling on cell viability: U-118 cells cultured in 96-well plates were labeled for 60 minutes with Qtracker® 565, 655, and 705 Kits. CellTiter 96 Non-Radioactive Cell Proliferation Assay (Promega) was performed on those cells 24 hours after labeling. The enzyme activity of unlabeled cells was used for normalization.

Table 2. Fluorescence excitation and emission maxima for Qtracker® Cell Labeling Kits.

Product	Catalog no.	Emission (nm)	Excitation (nm)
Qtracker® 525 Cell Labeling Kit	Q25041MP	525	405–485
Qtracker® 565 Cell Labeling Kit	Q25031MP	565	405–525
Qtracker® 585 Cell Labeling Kit	Q25011MP	585	405–545
Qtracker® 605 Cell Labeling Kit	Q25001MP	605	405–565
Qtracker® 625 Cell Labeling Kit	A10198	625	405–585
Qtracker® 655 Cell Labeling Kit	Q25021MP	655	405–615
Qtracker® 705 Cell Labeling Kit	Q25061MP	705	405–665
Qtracker® 800 Cell Labeling Kit	Q25071MP	800	405–760

See Figure 5 for Qdot® nanocrystal excitation and emission spectra.

Cell Labeling with Qtracker® Cell Labeling Kits

Labeling efficiency of your cells with the Qtracker® Cell Labeling Kit of your choice (Table 2) can be tested using the basic protocols for suspension or cultured adherent cells described below. Post-labeling, researchers have demonstrated a wide variety of applications for Qtracker® labeled cells, including cell co-culture and cell assembly into heterotypic assemblies,⁶ multilineage differentiation,⁷ transdifferentiation versus cell fusion,⁸ embryonic and mesenchymal stem cell tracking,⁹⁻¹⁰ and cell migration dynamics.¹²

Before You Begin

Note We recommend that you read the entire protocol before starting. For additional information, visit <http://probes.invitrogen.com/products/qdot>.

Experimental Protocol for Labeling Suspension Cells

The basic protocols below have been tested using a limited number of representative cell types (CHO, HeLa, U-118, 3T3, HUVEC, and Jurkat cells). Optimization may be required for optimal labeling of your cells based on your initial results. For example, in step 1.4 below, longer incubation times (2–24 hours) can be used for nanocrystal loading depending on experimental conditions and cell type. As part of optimization, we recommend performing cell viability tests (see Figure 4) using a standard cell proliferation assay method such as Vybrant® MTT Cell Proliferation Assay Kit (Invitrogen Cat. no. V13154).

Materials Required but Not Provided

- Mammalian suspension cells of choice
- Complete growth medium for the cell type used
- Microcentrifuge tubes

Qtracker® Cell Labeling Kit Protocol for Suspension Cells

- 1.1 To prepare 10 nM labeling solution, pre-mix 1 μ L each of Qtracker® Component A and Component B in a 1.5 mL microcentrifuge tube. Incubate for 5 minutes at room temperature and proceed immediately to Step 1.2.

Note: The working concentration of the Qtracker® label is typically in the range of 2 nM to 15 nM depending on the cell type and application. Prepare dilutions based on the 2 μ M concentration of Qtracker® Component A. Scale volumes as appropriate for the number of suspension cell samples to be treated.

- 1.2 Add 0.2 mL of fresh complete growth medium to the tube and vortex for 30 seconds.
- 1.3 Add 1×10^6 cells (from a cell suspension at $\sim 1 \times 10^7$ cells/mL in growth medium) to the tube containing the labeling solution.
- 1.4 Incubate the sample at 37°C for 45–60 minutes.
- 1.5 Wash cells twice with complete growth medium.
- 1.6 Visualize labeled live cells using any suitable fluorescence microscopy method or flow cytometry with appropriate filters (see Table 2 for fluorescence excitation/emission details).

Experimental Protocol for Labeling Adherent Cells

Materials Required but Not Provided

- Cell lines such as: HeLa cells (ATCC no. CCL-2) or U-118 cells (ATCC no. HTB-15)
- 8-well Lab-Tek chambered cover glass system or sterile coverslips suitable for subculturing cells in Petri dishes
- 75 cm² cell culture flask
- Optional: freshly-made 3.7% formaldehyde fixative solution (mix 1 mL 37–40% formaldehyde with 9 mL PBS)
- **ATCC medium for HeLa:**
Minimum essential medium (Eagle) with 2 mM L-glutamine and Earle's BSS adjusted to contain 1.5 g/L sodium bicarbonate, 0.1 mM non-essential amino acids, and 1.0 mM sodium pyruvate, 10% fetal bovine serum.
- **ATCC medium for U-118:**
Dulbecco's modified Eagle's medium with 4 mM L-glutamine adjusted to contain 1.5 g/L sodium bicarbonate and 4.5 g/L glucose, 10% fetal bovine serum

Qtracker[®] Cell Labeling Kit Protocol for Adherent Cells

Subculture Cells

- 2.1 Subculture HeLa or U-118 cells from 75 cm² cell culture flasks in 8-well Lab-Tek chambered coverglass system at a density of 2×10^4 cells per well (cell density may vary if using a different size plate). Cells subcultured on coverslips in culture plates and grown to similar confluence can also be utilized for small numbers of tests.
- 2.2 Incubate the cells in a 37°C, 5% CO₂ incubator overnight.

Labeling Procedure

- 3.1 To prepare 10 nM labeling solution, pre-mix 1 µL each of Qtracker[®] Component A and Component B in a 1.5 mL microcentrifuge tube. Incubate for 5 minutes at room temperature and proceed immediately to Step 3.2.

Note: The working concentration of the Qtracker[®] label is typically in the range of 2 nM to 15 nM depending on the cell type and application. Prepare dilutions based on the 2 µM concentration of Qtracker[®] Component A. Scale volumes as appropriate for the number of cell samples to be treated.
- 3.2 Add 0.2 mL of fresh complete growth medium to the tube and vortex for 30 seconds.
- 3.3 Add 0.2 mL of labeling solution to the well with cells. For labeling cells grown on coverslips, pipet ~0.15 mL labeling solution directly onto coverslips kept in a 60 mm Petri dish and cover.
- 3.4 Incubate at 37°C for 45–60 minutes.
- 3.5 Wash cells twice with complete growth medium.

Note: If desired, fix labeled cells at this point by washing 3 times with PBS, incubating with 3.7% formaldehyde in PBS for 15 minutes at room temperature, and washing 3 times post-fixation in PBS prior to imaging.
- 3.6 Visualize labeled cells using any suitable fluorescence microscopy method or flow cytometry with appropriate filters (see Table 2 for fluorescence excitation/emission details).

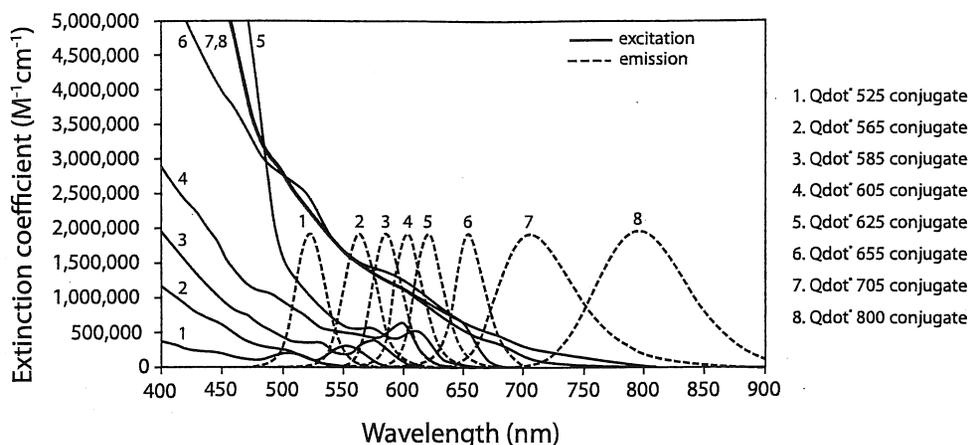


Figure 5. Typical absorption and emission spectra of Qdot[®] 525 conjugate (1), Qdot[®] 565 conjugate (2), Qdot[®] 585 conjugate (3), Qdot[®] 605 conjugate (4), Qdot[®] 625 conjugate (5), Qdot[®] 655 conjugate (6), Qdot[®] 705 conjugate (7), Qdot[®] 800 conjugate (8).

Stem Cell Results

We have demonstrated the usefulness of a co-culture/cell separation strategy in stem cell work by labeling mouse embryonic fibroblasts (MEFs) with the Qtracker[®] 655 Cell Labeling Kit and culturing the cells with SA2p12 hESCs or BG1vp22 human embryonic stem cells (hESCs). Flow cytometry analysis of MEF cells transfected with Qtracker[®] 655 showed 97% efficiency in labeling (Figure 6). MEFs labeled with Qtracker[®] are easily discriminated from colonies of BG1vp22 hESCs (Figure 7) and suspensions of SA2p12 hESCs (Figure 8). In fact, hESC colonies appear to exclude the feeder cells, rather than grow on top of them. Furthermore, excellent separation of hESCs and MEFs was obtained by flow cytometry (Figure 9). These findings illustrate the usefulness of Qtracker[®] kits for labeling live feeder cells and discriminating them from co-cultured hESCs.

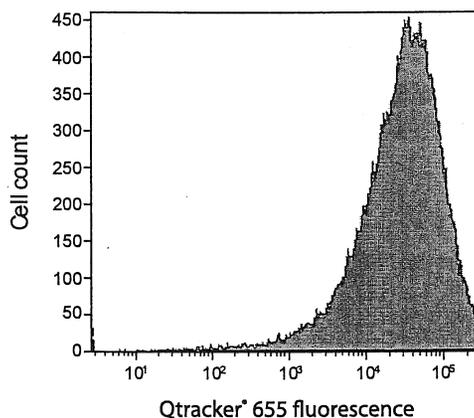


Figure 6. Flow cytometry analysis of MEFs transfected with Qtracker[®] 655 Cell Labeling Kit shows 97% labeling efficiency. Samples were analyzed on a FACS Canto[™] (Becton Dickinson).

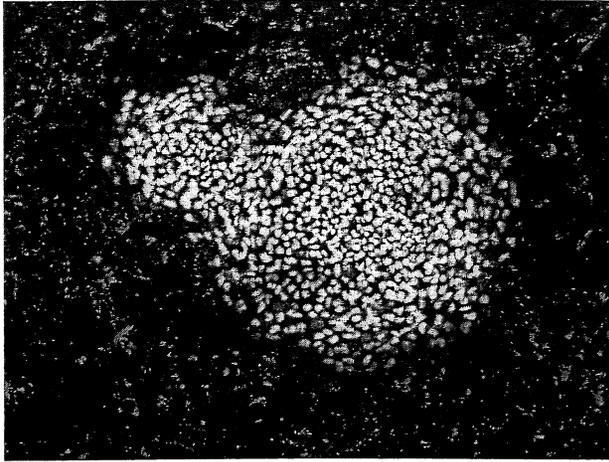


Figure 7. Mouse embryonic fibroblasts (MEFs) labeled with Qtracker® kits are easily discriminated from colonies of BG1vp22 human embryonic stem cells (hESCs). Colony of Oct 4-expressing BG1vp22 hESCs (Green; labeled with Alexa Fluor® 488 goat anti-rabbit IgG – Invitrogen Cat. no. A-11034) co-cultured with Qtracker® 655 labeled MEFs (Red) and counterstained with DAPI (Blue). Samples were imaged using a Nikon Eclipse TE300. Image capture was done using a Nuance™ multispectral imaging system.

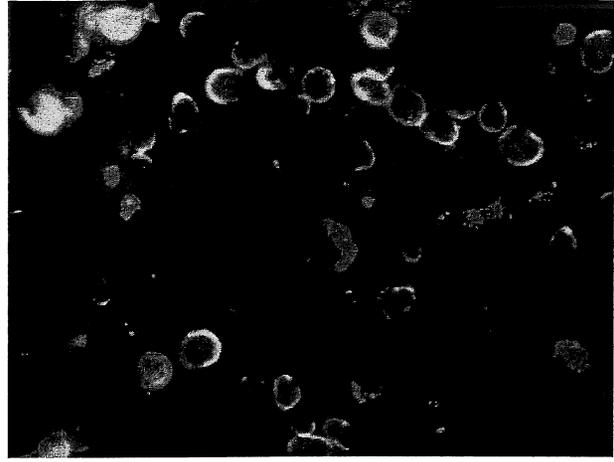


Figure 8. Discrimination of MEFs labeled with Qtracker nanocrystals from suspensions of SA2p12 hESCs. Suspension of Tra-1-81 expressing SA2p12 hESCs (Green; labeled with Alexa Fluor® 488 goat anti-mouse IgM – Invitrogen Cat. no. A-21042) co-cultured with Qtracker® 655 labeled MEFs (Red). Samples were imaged using a Nikon Eclipse TE300. Image capture was done using a Nuance™ multispectral imaging system.

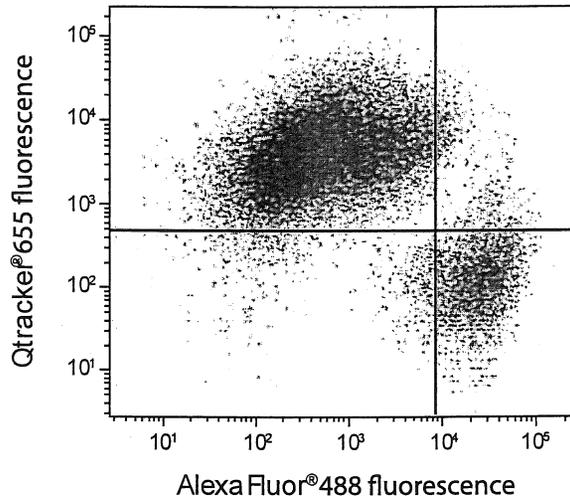


Figure 9. Separation of hESCs and MEFs by flow cytometry: Flow cytometry separation of SSEA4 expressing BG01vp29 cells (labeled with Alexa Fluor® 488 goat anti-mouse IgG₃ (γ3), Invitrogen Cat. no. A21151) from Qtracker® 655 labeled cells. Due to the brightness of Qtracker® 655 labeled signals compared to Alexa Fluor® 488 dye signals, TransFluoSpheres® streptavidin-labeled microspheres, 0.04 μm (488/465) (Invitrogen Cat. no. T10711) was used to better match scales.

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10. J Nanobiotechnology 5, 9 (2007);
11. Stem Cells 25, 2128 (2007);
12. Cytotechnology 51, 7 (2006).

Product List Current prices are available from www.invitrogen.com or from our Customer Service Department

Cat. no.	Product Name	Unit Size
Q25041MP	Qtracker [®] 525 Cell Labeling Kit.....	1 kit
Q25031MP	Qtracker [®] 565 Cell Labeling Kit.....	1 kit
Q25011MP	Qtracker [®] 585 Cell Labeling Kit.....	1 kit
Q25001MP	Qtracker [®] 605 Cell Labeling Kit.....	1 kit
A10198	Qtracker [®] 625 Cell Labeling Kit.....	1 kit
Q25021MP	Qtracker [®] 655 Cell Labeling Kit.....	1 kit
Q25061MP	Qtracker [®] 705 Cell Labeling Kit.....	1 kit
Q25071MP	Qtracker [®] 800 Cell Labeling Kit.....	1 kit

Related Products

A11034	Alexa Fluor [®] 488 goat anti-rabbit IgG.....	0.5 mL
A21042	Alexa Fluor [®] 488 goat anti-mouse IgM.....	250 µL
A21151	Alexa Fluor [®] 488 goat anti-mouse IgG ₃ (γ3).....	250 µL
T10711	TransFluoSpheres [®] streptavidin-labeled microspheres.....	0.4 mL
V13154	Vybrant [®] MTT Cell Proliferation Assay Kit.....	1 kit

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Brighter, Longer-lasting Signal and Better Cell Visualization with Qmount™ Qdot® Mounting Media and Qnuclear™ Deep Red Stain

Introduction

Designed for use with cells labeled with Qdot® nanocrystals, the recent releases of Qmount™ Qdot® Mounting Media and Qnuclear™ Deep Red Stain form a pair of important new tools that further enables the utility of Molecular Probes® Qdot® nanocrystal technology for fluorescence microscopy.

Qmount™ Qdot® Mounting Media (Cat. no. Q10336)

Qdot® fluorescence is susceptible to chemical quenching which represents a significant problem with conventional mounting media. Qmount™ Qdot® Mounting Media is a non-aqueous, permanent mountant optimized for performing microscopy with samples labeled with Qdot® nanocrystals. Unlike other mountants, the Qmount™ Qdot® Mounting Media causes no significant loss of the Qdot® nanocrystals' fluorescence, both initially and over the course of several months (Figure 1). This mounting media offers excellent compatibility with all eight Qdot® nanocrystals (Qdot® 525, 565, 585, 605, 625, 655, 705, and 800), their conjugates, and Qnuclear™ Deep Red stain (Cat. no. Q10363), making it an especially valuable tool for multicolor Qdot® nanocrystal imaging applications. Although optimal for use with Qdot® nanocrystals, this mounting medium is not recommended for use with most standard organic dyes or fluorescent proteins.

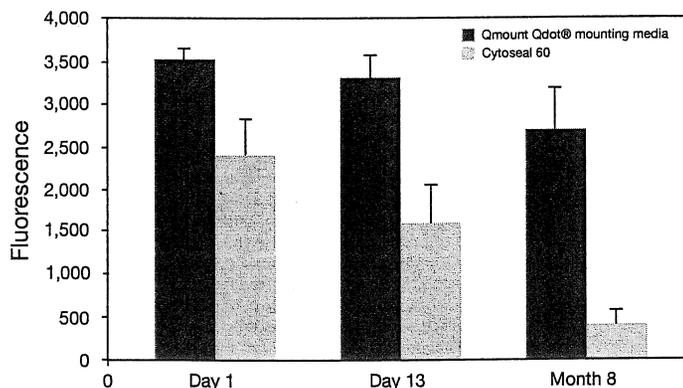


Figure 1. Comparison of fluorescently labeled mammalian cells in Qmount™ Qdot® Mounting Media and Cytoseal™ 60 reagent, imaged on day 1 and day 13, and after 8 months. Qmount™ Qdot® Mounting Media enhances the photostability of Qdot® nanocrystals, both initially and over time. Human carcinoma (HeLa) cells labeled with mouse anti-OxPhos Complex V inhibitor protein IgG (Cat. no. A21355) and Qdot® 605 conjugated goat anti-mouse IgG (Cat. no. Q11001MP) were mounted with Qmount™ or Cytoseal™ 60 mountant (Thermo Scientific) and illuminated using a 100-watt Hg-arc lamp.

Qnuclear™ Deep Red Stain (Cat. no. Q10363)

The new Qnuclear™ Deep Red stain is a nuclear counterstain specifically designed for use with cells labeled with Qdot® 525, 565, 585, 605, 625, and 655 nanocrystals, providing bright and photostable nuclear counterstaining for cell identification and multiplex imaging with no overlap into the excitation wavelengths of Qdot® nanocrystals. With excitation and emission maxima of 640 and 663 nm, respectively, this counterstain can be visualized with standard fluorescence microscopy filter sets (Figure 2).

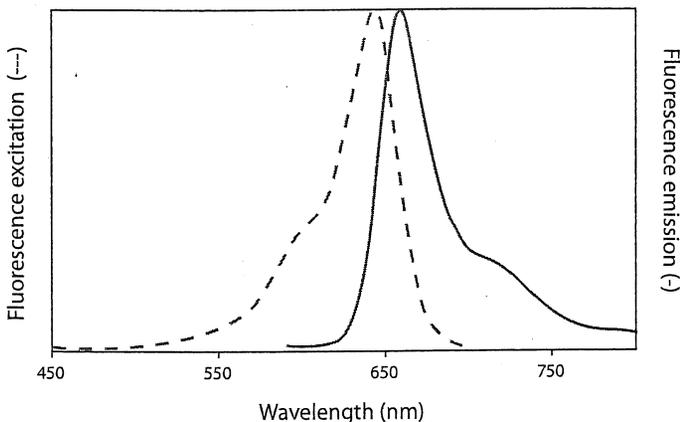


Figure 2. Fluorescence excitation and emission spectra for Qnuclear™ Deep Red Stain.

Product List Current prices may be obtained from our website or from our Customer Service Department.

Cat. no.	Product Name	Unit Size
Q10336	Qmount™ Qdot® Mounting Media	3 × 2 mL
Q10363	Qnuclear™ Deep Red Stain	100 µL

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Qtracker® Cell Labeling Kits

Catalog nos. A10198, Q25001MP, Q25011MP, Q25021MP, Q25031MP, Q25041MP, Q25061MP, Q25071MP

Table 1. Contents and storage information.

Material	Amount	Concentration	Storage	Stability
Qtracker® nanocrystals, Component A	100 µL	2 µM in 50 mM borate buffer, pH 8.3	<ul style="list-style-type: none"> • 2–6°C • DO NOT FREEZE 	When stored as directed the product is stable for at least 6 months.
Qtracker® carrier, Component B	100 µL	Phosphate buffered saline (PBS), pH 7.2		

Approximate fluorescence excitation and emission maxima: See Table 2.

Introduction

Qtracker® Cell Labeling Kits are designed for loading cells grown in culture with highly fluorescent Qdot® nanocrystals. Once inside the cells, Qtracker® labels provide intense, stable fluorescence that can be traced through several generations, and are not transferred to adjacent cells in a population.

Qtracker® Cell Labeling Kits are available in seven colors—525 nm, 565 nm, 585 nm, 605 nm, 625 nm, 655 nm, 705 nm, or 800 nm emission—and are excellent tools for long-term tracking or imaging studies of live cells, including migration, motility, morphology, and other cell function assays.

The Qtracker® Cell Labeling Kits use a custom targeting peptide^{1,2} to deliver Qdot® nanocrystals into the cytoplasm of live cells. Cytoplasmic delivery by this mechanism is not mediated by a specific enzyme; therefore, no cell-type specificity has been observed. Delivery is typically accomplished in less than 1 hour. Qdot® nanocrystals delivered by the Qtracker® Cell Labeling Kits are compatible with serum-sensitive cells; intense fluorescence is maintained in complex cellular environments and under various biological conditions including changes in intracellular pH, temperature, and metabolic activity. Furthermore, autofluorescence commonly observed in cells or tissues can be avoided using Qtracker® 655, 705, or 800 Kits.

Features and Applications

Using Qtracker® Cell Labeling Kits, you can observe labeled cells using extensive continuous illumination, without the photobleaching and degradation problems often associated with organic dyes.³⁻⁵ Qtracker® labels are distributed in vesicles in the cytoplasm (Figure 1), and are inherited by daughter cells for at least six generations. Fluorescence from the Qtracker® labels can be seen up to a week after delivery in some cell lines. Long-term cellular retention makes Qtracker® Cell Labeling Kits ideal for studying cell motility (Figure 2), migration,

differentiation, morphology, and many other cellular function studies.^{3,4} Qtracker® labels do not leak out of intact cells to be taken up by adjacent cells in the population (Figure 3).

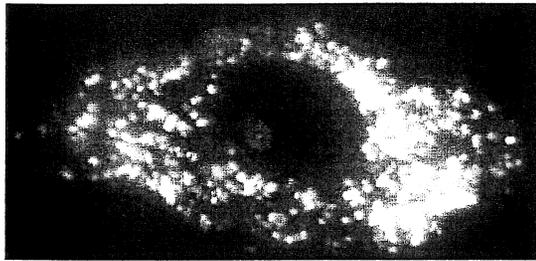


Figure 1. Distribution of Qtracker® labels in the cytoplasm: HeLa cells labeled with the Qtracker® 655 Cell Labeling Kit were observed with a Leica TCS SP2 confocal microscopy to see the distribution of the Qtracker® reagent in the cytoplasm (excitation at 488 nm).

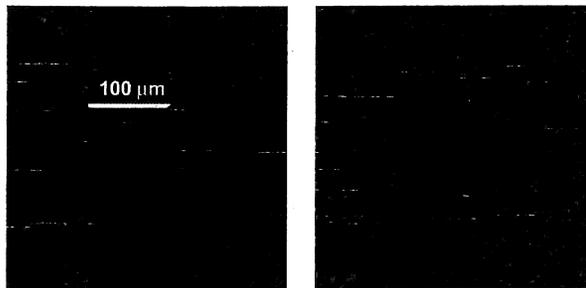


Figure 2. Motility of HeLa cells: A monolayer of HeLa cells was labeled with the Qtracker® 655 Cell Labeling Kit. The gap was made by scratching with a 200 μ L pipette tip (~1 mm) and imaged using a Leica TCS SP2 confocal microscope (excitation at 488 nm; 10X objective). The cells moved to fill the gap and retained normal motility.

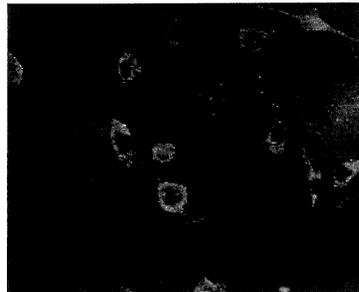


Figure 3. U-118 and HeLa cells were labeled with Qtracker® 565 and 655 Cell Labeling Kits, respectively, and co-cultured for 24 hours. The image was captured using a Leica TCS SP2 confocal microscope. Both colors are easily resolved, well retained, and well segregated in their respective cell lines.

Imaging Platforms

Qtracker® reagent-labeled live cells can be easily monitored on a variety of platforms, including flow cytometry, fluorescence/confocal microscopy, fluorescence microplate readers, and high-content imaging systems.

Cytotoxicity and Viability Studies

The cytotoxicity of the materials use in Qtracker® Cell Labeling Kits has been tested in a variety of cell lines including CHO, HeLa, U-118, 3T3, HUVEC, and Jurkat cells. Labeling with Qtracker® Cell Labeling Kits appears to exert minimal impact on cellular surface marker expression, cell proliferation, cellular enzyme activity, and cell motility; no effect on the CD3 expression level of Jurkat cells

was observed following labeling with the Qtracker® Cell Labeling Kit.

The effect of Qdot® nanocrystal loading on cellular viability has been examined using cell proliferation and cellular enzyme activity measurements (Figure 4). The results indicate that labeling with the Qtracker® Cell Labeling Kit has no significant effect on cell proliferation and cellular enzyme activity.

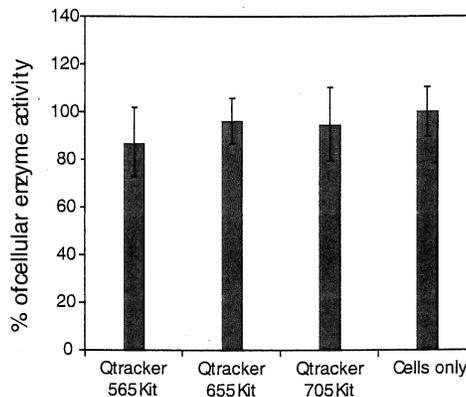


Figure 4. Effect of Qtracker® labeling on cell viability: U-118 cells cultured in 96-well plates were labeled for 60 minutes with Qtracker® 565, 655, and 705 Kits. CellTiter 96 Non-Radioactive Cell Proliferation Assay (Promega) was performed on those cells 24 hours after labeling. The enzyme activity of unlabeled cells was used for normalization.

Table 2. Fluorescence excitation and emission maxima for Qtracker® Cell Labeling Kits.

Product	Catalog no.	Emission (nm)	Excitation (nm)
Qtracker® 525 Cell Labeling Kit	Q25041MP	525	405–485
Qtracker® 565 Cell Labeling Kit	Q25031MP	565	405–525
Qtracker® 585 Cell Labeling Kit	Q25011MP	585	405–545
Qtracker® 605 Cell Labeling Kit	Q25001MP	605	405–565
Qtracker® 625 Cell Labeling Kit	A10198	625	405–585
Qtracker® 655 Cell Labeling Kit	Q25021MP	655	405–615
Qtracker® 705 Cell Labeling Kit	Q25061MP	705	405–665
Qtracker® 800 Cell Labeling Kit	Q25071MP	800	405–760

See Figure 5 for Qdot® nanocrystal excitation and emission spectra.

Cell Labeling with Qtracker® Cell Labeling Kits

Labeling efficiency of your cells with the Qtracker® Cell Labeling Kit of your choice (Table 2) can be tested using the basic protocols for suspension or cultured adherent cells described below. Post-labeling, researchers have demonstrated a wide variety of applications for Qtracker® labeled cells, including cell co-culture and cell assembly into heterotypic assemblies,⁶ multilineage differentiation,⁷ transdifferentiation versus cell fusion,⁸ embryonic and mesenchymal stem cell tracking,⁹⁻¹⁰ and cell migration dynamics.¹²

Before You Begin

Note We recommend that you read the entire protocol before starting. For additional information, visit <http://probes.invitrogen.com/products/qdot>.

Experimental Protocol for Labeling Suspension Cells

The basic protocols below have been tested using a limited number of representative cell types (CHO, HeLa, U-118, 3T3, HUVEC, and Jurkat cells). Optimization may be required for optimal labeling of your cells based on your initial results. For example, in step 1.4 below, longer incubation times (2–24 hours) can be used for nanocrystal loading depending on experimental conditions and cell type. As part of optimization, we recommend performing cell viability tests (see Figure 4) using a standard cell proliferation assay method such as Vybrant® MTT Cell Proliferation Assay Kit (Invitrogen Cat. no. V13154).

Materials Required but Not Provided

- Mammalian suspension cells of choice
- Complete growth medium for the cell type used
- Microcentrifuge tubes

Qtracker® Cell Labeling Kit Protocol for Suspension Cells

- 1.1 To prepare 10 nM labeling solution, pre-mix 1 μ L each of Qtracker® Component A and Component B in a 1.5 mL microcentrifuge tube. Incubate for 5 minutes at room temperature and proceed immediately to Step 1.2.

Note: The working concentration of the Qtracker® label is typically in the range of 2 nM to 15 nM depending on the cell type and application. Prepare dilutions based on the 2 μ M concentration of Qtracker® Component A. Scale volumes as appropriate for the number of suspension cell samples to be treated.

- 1.2 Add 0.2 mL of fresh complete growth medium to the tube and vortex for 30 seconds.
- 1.3 Add 1×10^6 cells (from a cell suspension at $\sim 1 \times 10^7$ cells/mL in growth medium) to the tube containing the labeling solution.
- 1.4 Incubate the sample at 37°C for 45–60 minutes.
- 1.5 Wash cells twice with complete growth medium.
- 1.6 Visualize labeled live cells using any suitable fluorescence microscopy method or flow cytometry with appropriate filters (see Table 2 for fluorescence excitation/emission details).

Experimental Protocol for Labeling Adherent Cells

Materials Required but Not Provided

- Cell lines such as: HeLa cells (ATCC no. CCL-2) or U-118 cells (ATCC no. HTB-15)
- 8-well Lab-Tek chambered cover glass system or sterile coverslips suitable for subculturing cells in Petri dishes
- 75 cm² cell culture flask
- Optional: freshly-made 3.7% formaldehyde fixative solution (mix 1 mL 37–40% formaldehyde with 9 mL PBS)
- **ATCC medium for HeLa:**
Minimum essential medium (Eagle) with 2 mM L-glutamine and Earle's BSS adjusted to contain 1.5 g/L sodium bicarbonate, 0.1 mM non-essential amino acids, and 1.0 mM sodium pyruvate, 10% fetal bovine serum.
- **ATCC medium for U-118:**
Dulbecco's modified Eagle's medium with 4 mM L-glutamine adjusted to contain 1.5 g/L sodium bicarbonate and 4.5 g/L glucose, 10% fetal bovine serum

Qtracker[®] Cell Labeling Kit Protocol for Adherent Cells

Subculture Cells

- 2.1 Subculture HeLa or U-118 cells from 75 cm² cell culture flasks in 8-well Lab-Tek chambered coverglass system at a density of 2×10^4 cells per well (cell density may vary if using a different size plate). Cells subcultured on coverslips in culture plates and grown to similar confluence can also be utilized for small numbers of tests.
- 2.2 Incubate the cells in a 37°C, 5% CO₂ incubator overnight.

Labeling Procedure

- 3.1 To prepare 10 nM labeling solution, pre-mix 1 µL each of Qtracker[®] Component A and Component B in a 1.5 mL microcentrifuge tube. Incubate for 5 minutes at room temperature and proceed immediately to Step 3.2.
Note: The working concentration of the Qtracker[®] label is typically in the range of 2 nM to 15 nM depending on the cell type and application. Prepare dilutions based on the 2 µM concentration of Qtracker[®] Component A. Scale volumes as appropriate for the number of cell samples to be treated.
- 3.2 Add 0.2 mL of fresh complete growth medium to the tube and vortex for 30 seconds.
- 3.3 Add 0.2 mL of labeling solution to the well with cells. For labeling cells grown on coverslips, pipet ~0.15 mL labeling solution directly onto coverslips kept in a 60 mm Petri dish and cover.
- 3.4 Incubate at 37°C for 45–60 minutes.
- 3.5 Wash cells twice with complete growth medium.
Note: If desired, fix labeled cells at this point by washing 3 times with PBS, incubating with 3.7% formaldehyde in PBS for 15 minutes at room temperature, and washing 3 times post-fixation in PBS prior to imaging.
- 3.6 Visualize labeled cells using any suitable fluorescence microscopy method or flow cytometry with appropriate filters (see Table 2 for fluorescence excitation/emission details).

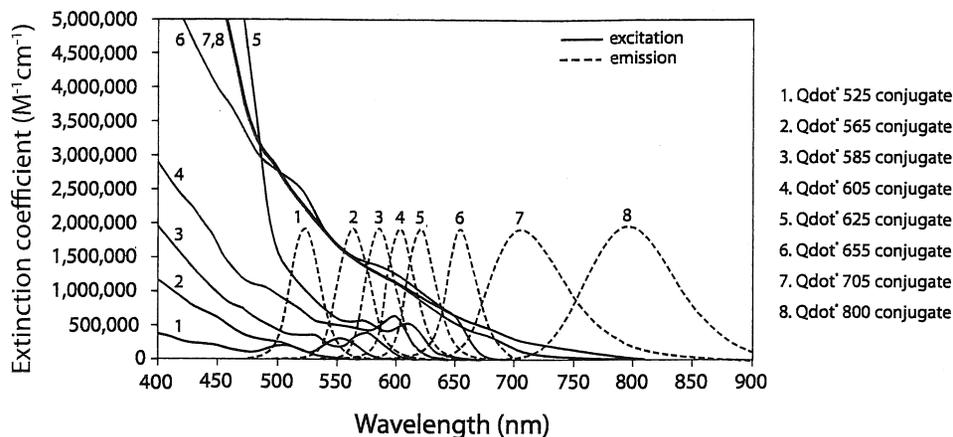


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Stem Cell Results

We have demonstrated the usefulness of a co-culture/cell separation strategy in stem cell work by labeling mouse embryonic fibroblasts (MEFs) with the Qtracker[®] 655 Cell Labeling Kit and culturing the cells with SA2p12 hESCs or BG1vp22 human embryonic stem cells (hESCs). Flow cytometry analysis of MEF cells transfected with Qtracker[®] 655 showed 97% efficiency in labeling (Figure 6). MEFs labeled with Qtracker[®] are easily discriminated from colonies of BG1vp22 hESCs (Figure 7) and suspensions of SA2p12 hESCs (Figure 8). In fact, hESC colonies appear to exclude the feeder cells, rather than grow on top of them. Furthermore, excellent separation of hESCs and MEFs was obtained by flow cytometry (Figure 9). These findings illustrate the usefulness of Qtracker[®] kits for labeling live feeder cells and discriminating them from co-cultured hESCs.

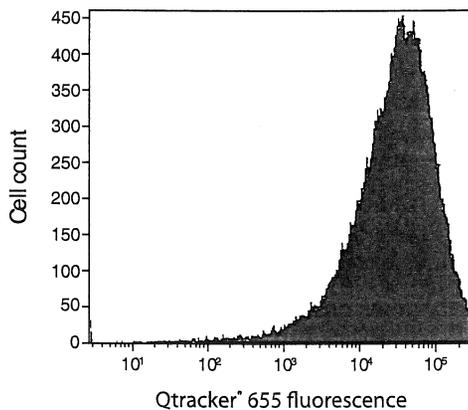


Figure 6. Flow cytometry analysis of MEFs transfected with Qtracker[®] 655 Cell Labeling Kit shows 97% labeling efficiency. Samples were analyzed on a FACS Canto[™] (Becton Dickinson).

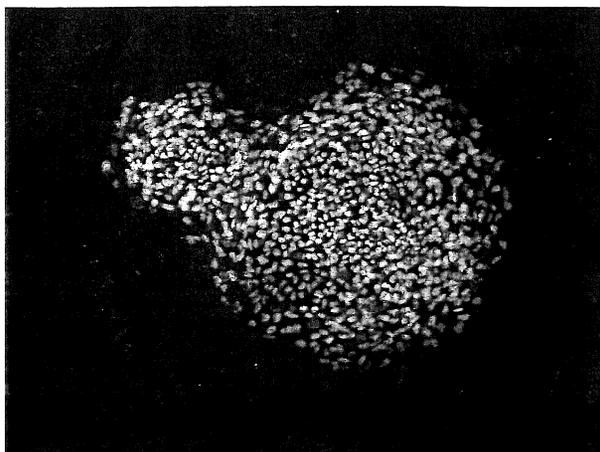


Figure 7. Mouse embryonic fibroblasts (MEFs) labeled with Qtracker® kits are easily discriminated from colonies of BG1vp22 human embryonic stem cells (hESCs). Colony of Oct 4-expressing BG1vp22 hESCs (Green; labeled with Alexa Fluor® 488 goat anti-rabbit IgG – Invitrogen Cat. no. A-11034) co-cultured with Qtracker® 655 labeled MEFs (Red) and counterstained with DAPI (Blue). Samples were imaged using a Nikon Eclipse TE300. Image capture was done using a Nuance™ multispectral imaging system.

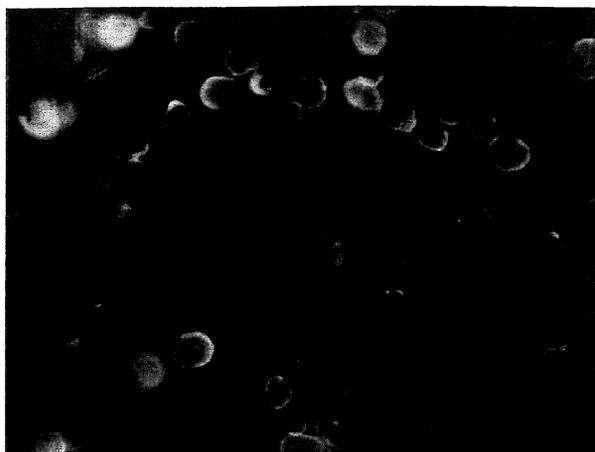


Figure 8. Discrimination of MEFs labeled with Qtracker nanocrystals from suspensions of SA2p12 hESCs. Suspension of Tra-1-81 expressing SA2p12 hESCs (Green; labeled with Alexa Fluor® 488 goat anti-mouse IgM – Invitrogen Cat no. A-21042) co-cultured with Qtracker® 655 labeled MEFs (Red). Samples were imaged using a Nikon Eclipse TE300. Image capture was done using a Nuance™ multispectral imaging system.

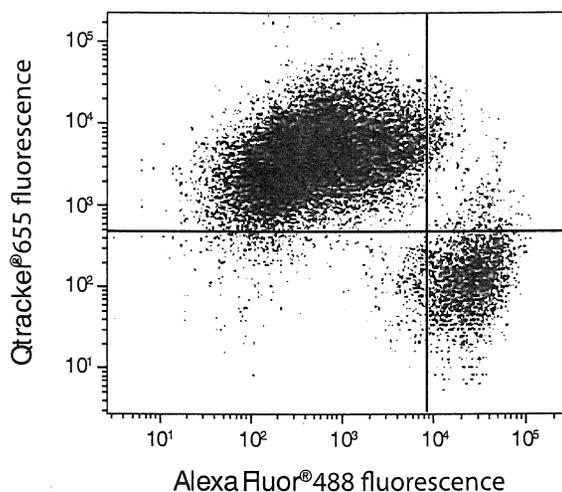


Figure 9. Separation of hESCs and MEFs by flow cytometry: Flow cytometry separation of SSEA4 expressing BG01vp29 cells (labeled with Alexa Fluor® 488 goat anti-mouse IgG₃ (γ3), Invitrogen Cat. no. A21151) from Qtracker® 655 labeled cells. Due to the brightness of Qtracker® 655 labeled signals compared to Alexa Fluor® 488 dye signals, TransFluoSpheres® streptavidin-labeled microspheres, 0.04 μm (488/465) (Invitrogen Cat. no. T10711) was used to better match scales.

References

1. Curr Protein Pept Sci 4, 87 (2003);
2. J Am Chem Soc 124, 368 (2002);
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4. Biochem Biophys Res Comm 302, 496 (2003);
5. Nano Lett 4, 2019 (2004);
6. Am J Pathology 168, 1793 (2006);
7. Stem Cells 25, 2760 (2007);
8. Arterioscler Thromb Vasc Biol 25, 1388 (2005);
9. BMC Biotechnol 7,67 (2007);
10. J Nanobiotechnology 5, 9 (2007);
11. Stem Cells 25, 2128 (2007);
12. Cytotechnology 51, 7 (2006).

Product List Current prices are available from www.invitrogen.com or from our Customer Service Department

Cat. no.	Product Name	Unit Size
Q25041MP	Qtracker [®] 525 Cell Labeling Kit.....	1 kit
Q25031MP	Qtracker [®] 565 Cell Labeling Kit.....	1 kit
Q25011MP	Qtracker [®] 585 Cell Labeling Kit.....	1 kit
Q25001MP	Qtracker [®] 605 Cell Labeling Kit.....	1 kit
A10198	Qtracker [®] 625 Cell Labeling Kit.....	1 kit
Q25021MP	Qtracker [®] 655 Cell Labeling Kit.....	1 kit
Q25061MP	Qtracker [®] 705 Cell Labeling Kit.....	1 kit
Q25071MP	Qtracker [®] 800 Cell Labeling Kit.....	1 kit
Related Products		
A11034	Alexa Fluor [®] 488 goat anti-rabbit IgG.....	0.5 mL
A21042	Alexa Fluor [®] 488 goat anti-mouse IgM.....	250 µL
A21151	Alexa Fluor [®] 488 goat anti-mouse IgG ₃ (γ3).....	250 µL
T10711	TransFluoSpheres [®] streptavidin-labeled microspheres.....	0.4 mL
V13154	Vybrant [®] MTT Cell Proliferation Assay Kit.....	1 kit

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Attachment 3:

Section D-F for Nano Silver

Section D: Nanomaterial Chemical and Physical Properties			
Product / Production Information (Based on five Stanford University academic research laboratories)			
Nano Chemical Name:		Nano Silver	
Commercial Name(s):		N/A (synthesized by four SU academic research laboratories) Silver Nanowires (purchased by one lab)	
Annual Production Volume:		~13 g (synthesized by four SU academic research laboratories) 1 g (purchased by one lab)	
Production Method(s):		Reduction of Silver salt; Chemical Synthesis; Polyol process Unknown (commercial product)	
Identification of the Supplier(s):		N/A (synthesized and used by SU academic research laboratories) Seashell Technology LLC	
Parameter		Value / Range ¹ (include units)	Name of Analytical Method(s) ²
Physical Properties			
Shape (Morphology)		Spheres, Nanowires, Nanocubes, Cylinders	Atomic Force Microscopy; Scanning Electron Microscopy, Transmission Electron Microscopy
Density		~10.5 g/cm ³	Theoretical
Surface Area		<ul style="list-style-type: none"> ~57 m²/g (spheres) 2-95 μm² (cylinders) Unknown (others) 	<ul style="list-style-type: none"> By calculation based on monodispersed particles Scanning Electron Microscopy Unknown
Particle Size Distribution	Air	<ul style="list-style-type: none"> 7-40 μm (cylinders) Unknown (others) 	<ul style="list-style-type: none"> Scanning Electron Microscopy Unknown
	Liquid	<ul style="list-style-type: none"> 3-35 nm (spheres) 30-100 nm diameter x 3-20 μm length (nanowires) 7-40 μm (cylinders) 	<ul style="list-style-type: none"> Atomic Force Microscopy, Transmission Electron Microscopy Scanning Electron Microscopy, Transmission Electron Microscopy Scanning Electron Microscopy
	Solid / Powder	<ul style="list-style-type: none"> 7-35 nm (spheres) 7-40 μm (cylinders) Unknown (others) 	<ul style="list-style-type: none"> Transmission Electron Microscopy, Scanning Electron Microscopy Transmission Electron Microscopy Scanning Electron Microscopy Unknown
Other (Specify)			
Chemical Properties			
Chemical Composition		Ag	Transmission Electron Microscopy, Inductively Coupled Plasma-Optical Emission Spectrometry, Energy-dispersive X-ray spectroscopy
Surface Modification (Coating, Functionalization)		Alkylamine, Polyvinylpyrrolidone	X-Ray Photoelectron Spectroscopy
Purity		Unknown	Unknown
Surface Charge		<ul style="list-style-type: none"> Negative (pH 2-12) (sphere) Unknown (others) 	<ul style="list-style-type: none"> Zetasizer (Zeta Potential Measurement) Unknown
Dispersion ³	Air	Unknown	Unknown
	Liquid	<ul style="list-style-type: none"> Monodispersed (spheres) Fully dispersed with sonication (nanowires, nanocubes) Unknown (cylinders) 	<ul style="list-style-type: none"> Visual examination, Dynamic Light Scattering Visual examination, Scanning Electron Microscopy Unknown

	Solid	<ul style="list-style-type: none"> • Agglomerate easily (spheres) • Unknown (others) 	<ul style="list-style-type: none"> • Dynamic Light Scattering • Unknown
Identifying and Determining Concentration of Nano Chemical, Its Metabolites, and Degradation Products in Specified Matrices Water, Air, Soil, Sediment, Sludge, Chemical Waste, Fish, Blood, Adipose Tissue, Urine, Other (Specify)		See Section F	
Solubility	Water Solubility	<ul style="list-style-type: none"> • 2-10% (sphere) • Soluble (nanowires, nanocubes) • Not Soluble (cylinder) 	<ul style="list-style-type: none"> • Inductively Coupled Plasma-Optical Emission Spectrometry • Visual examination • Supplier information
	Solubility in Organic Solvent	<ul style="list-style-type: none"> • ~200mg/ml (spheres) • Reported as Soluble and Not Soluble (nanowires) • Soluble (cylinder) 	<ul style="list-style-type: none"> • Visual examination • Visual examination • Supplier information
n-Octanol- Water Partition Coefficient		Unknown	Unknown
Stability and Reactivity	Flammability	<ul style="list-style-type: none"> • Same as Isopropanol (cylinder) • Not Flammable (others) 	<ul style="list-style-type: none"> • Supplier information • Based on bulk silver
	Explosiveness	<ul style="list-style-type: none"> • Same as Isopropanol (cylinder) • Not Explosive (others) 	<ul style="list-style-type: none"> • Supplier information • Based on bulk silver
	Oxidizing Properties	<ul style="list-style-type: none"> • Reacts with strong oxidizing agents (cylinders) • Unknown (others) 	<ul style="list-style-type: none"> • Supplier information • Unknown
	Oxidation Reduction Potential	<ul style="list-style-type: none"> • 0.8 V vs. SHE (nanowires) • Unknown (others) 	<ul style="list-style-type: none"> • Electrochemical Measurements • Unknown
	Storage Stability and Reactivity (Container Material)	<ul style="list-style-type: none"> • Stable in plastic (cylinders) • Stable (others) 	<ul style="list-style-type: none"> • Supplier information • Visual examination
	Stability to Thermal, Sunlight, and Metal(s)	<ul style="list-style-type: none"> • Unknown (spheres) • Will decompose at temperatures above 180°C and Degrades in light (nanowires) • Stable (cylinder) 	<ul style="list-style-type: none"> • Unknown (spheres) • Heated in oven; Visible observation • Heated in oven until melting at ~150°C (cylinder)

Section E: Globally Harmonized System Safety Data Sheet (SDS) or Materials Safety Data Sheet (MSDS)

Chemical Name	Source
Silver nanowires	Seashell Technology LLC

Section F: Describe the analytical test method(s) that you use or plan to use to sample, prepare and analyze a specific matrix (water, air, soil, sediment, sludge, chemical waste, fish, blood, adipose tissue, and urine) to determine the identity and concentration of each specified nanomaterial.

Nanomaterial	Purpose (sample, prepare, analyze)	Matrix
Silver nanoparticles	Analyze	Water
Method description		
Working with water solutions ranging from 1 ppb to 10000 ppm levels the solubility and environmental transformations are studies using Inductively Coupled Plasma-Optical Emission Spectrometry.		

MATERIAL SAFETY DATA SHEETS



Seashell Technology

Seashell Technology, LLC
3252 Holiday Court, Suite 115
La Jolla, CA 92037
858 638 0315
858 638 0376 (fax)
www.seashelltech.com

Material Safety Data Sheet (MSDS)

Section 1: Product and Company Identification

PRODUCT NAME: Silver NanoWires
SYNONYMS: Ag, Ag NanoWires, Ag NW, Silver, Silver Particles, Silver NanoRods
CAS NO.: 7440-22-4

MANUFACTURER: Seashell Technology, LLC
ADDRESS: 3252 Holiday Court, Suite 115
La Jolla, CA 92037

EMERGENCY PHONE: 858 750 9340
OTHER CALLS: 858 638 0315
FAX: 858 638 0376

Section 2: Composition/Information on Ingredients

INGREDIENTS	CAS No	PERCENT	EXPOSURE LIMITS
Silver	7440-22-4	.5 - 5	N/A
Isopropanol	87-83-0	95-99.5	400ppm (OSHA/PEL) 200ppm (ACGIH/TLV)

Section 3: Hazards Identification

Routes of Exposure:

Ingestion: Slightly toxic. May cause nausea, abdominal discomfort, vomiting, dizziness, and gastrointestinal irritation.

Skin Absorption: May cause skin irritation or ulceration.

Inhalation: May be harmful if inhaled. Material may be irritating to nasal septum, throat, mucous membranes and upper respiratory tract.

Eye Contact: May cause irritation including stinging, tearing, and redness. May cause corneal injury or blue-gray eyes.

Effects of Repeated Overexposure: Absorption of silver compounds by ingestion, inhalation or through broken skin can cause argyria, a permanent bluish-gray discoloration of the skin, conjunctiva and mucous membranes. Skin contact may aggravate an existing dermatitis.

Other Health Hazards: None currently known.

Section 4: First Aid Measures

Obtain medical attention for all cases of over-exposure.

Symptoms and effects: Headache, Dizziness, Nausea, Narcosis, Dryness of the skin. Ingestion may cause inebriation and coma. Irritation of the skin, eyes, and respiratory tract.
Ingestion: If conscious, wash out mouth with water. DO NOT induce vomiting. If rapid recovery does not occur, obtain medical attention. Give water to drink, providing patient is conscious.
Skin Absorption: Wash skin with soap and water for at least 15 minutes. If persistent irritation occurs, obtain medical attention. Remove contaminated clothing and shoes. Wash clothing before reuse. Thoroughly clean shoes before reuse.
Inhalation: Remove to fresh air. Give artificial respiration if not breathing. If breathing is difficult, oxygen may be given by qualified personnel. Obtain medical assistance if discomfort persists.
Eye Contact: Flush eyes with water for at least 15 minutes occasionally lifting lower and upper eyelids. Obtain medical attention.
Advice to physicians: Dermatitis may result from prolonged or repeated exposure. May cause central nervous system depression.

Section 5: Fire Fighting Measures

Fire/Explosive Properties
Flash Point: 53°F (12°C) Tag Closed Cup
Flammable Limits in Air: 2.0 - 12.7%
NFPA Rating: Health 1 Fire 3 Reactivity 1

Specific hazards: Hazardous combustion products may include carbon monoxide. The vapor is heavier than air, spreads along the ground and distant ignition is possible. May produce a floating fire hazard. Static ignition hazard can result from handling and use. Any very finely divided particles (ultra-fine powder) may burn in air. Combustion of silver powder may cause the release of toxic metal oxide fume.
Explosion: This material, like most materials in powder form, is capable of creating a dust explosion.
Extinguishing Media: Alcohol resistant foam, water spray or fog. Dry chemical powder, carbon dioxide, sand or earth may be used for small fires only. Sand or dry powder type specially designed for metal powder fires. Do not use water.
Unsuitable extinguishing media: Water in a jet.
Protective equipment: Full protective clothing and self contained breathing apparatus.
Other information: Keep adjacent containers cool by spraying with water. Use water spray to disperse vapors - re-ignition is possible in the event of a fire, wear full protective clothing and NIOSH-approved self contained breathing apparatus with full face piece operated in the pressure demand or other positive pressure mode.

Section 6: Accidental Release Measures

Personal precautions: In case of a leak or spill, evacuate area, shut off all sources of ignition and use non-sparking tools. Avoid contact with skin and eyes. Ventilate contaminated area thoroughly. Do not breathe vapor. Extinguish naked flames. No smoking. Avoid sparks. Evacuate the area of all non-essential personnel. Shut off leaks, if possible, without personal risk.
Personal protection: Wear PVC, neoprene, or nitrile rubber gloves, PVC one-piece suit with integral hood, and rubber, knee-length safety boots. Wear full face-piece respirator with organic vapor canister NPF 400. In a confined space, wear self-contained breathing apparatus open circuit type NPF 2000. For smaller spills, wear eye protection, self-contained breathing apparatus, boots, and protective gloves. Wear disposable coveralls and discard after use.
Environmental precautions: Prevent contamination of soil and water. Prevent from spreading or entering into drains, ditches, or rivers by using sand, earth, or other appropriate barriers
Clean-up methods - small spillage: Avoid raising dust. Absorb or contain liquid with sand, earth or spill control material. Shovel up and place in a labeled, sealable container for subsequent safe disposal. Put leaking containers in a labeled drum or overdrum. Flush contaminated area with plenty of water. Retain washings as contaminated waste.
Clean-up methods - large spillage: Avoid raising dust. Transfer to a labeled, sealable container for product recovery or safe disposal. Treat residues as for small spillage.

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Other information: Risk of explosion. Inform the emergency services if liquid enters surface water drains. Vapor may form an explosive mixture with air. See section 13 for information on disposal.

Section 7: Handling and Storage

Handling: observe all warnings and precautions listed for the product. Avoid prolonged or repeated contact with skin. Avoid contact with eyes. Wash thoroughly after handling. Extinguish any naked flames. Remove ignition sources. Avoid sparks. Do not smoke. Take precautionary measures against static discharges. Ground all equipment. Do not empty into drains.

Handling temperatures: Ambient.

Storage: Tanks should be fitted with a vapor recovery system. Vapors may settle in low or confined areas. Keep away from direct sunlight and other sources of heat or ignition. Do not smoke in storage areas. Keep container tightly closed and in a cool, dry, well-ventilated place. Vapors may collect in containers; treat empty containers as hazardous.

Storage temperatures: Ambient.

Product transfer: Take precautionary measures against static discharges. Earth all equipment. Avoid splash filling.

Recommended materials: For containers or container linings, use mild steel or stainless steel. If diluted with de-ionized water, steel containers may be unsuitable.

Section 8: Exposure Controls, Personal Protection

Occupational exposure standards:

Component name	Limit type	Value	Unit	Other information
Isopropyl alcohol	TLV/TWA	999	mg/m ³	(400ppm for 8 hours)
	STEL	1250	mg/m ³	(500ppm for 15 mins)
Silver	TLV/TWA	0.1	mg/m ³	
	PEL/TWA	0.01	mg/m ³	

Ventilation System: Use only in well-ventilated areas. A system of local and/or general exhaust is recommended to keep employee exposures below the Airborne Exposure Limits. Local exhaust ventilation is generally preferred because it can control the emission of the contaminant at its source, preventing dispersion of it into the general work area. Local ventilation is needed where vapors escape to the workplace air.

Respiratory protection: Respirators may be necessary when engineering and administrative controls do not adequately prevent exposures. Currently, there are no specific exposure limits for airborne exposures to engineered nanoparticles although occupational exposure limits exist for larger particles of similar chemical composition. The decision to use respiratory protection should be based on professional judgment that takes into account toxicity information, exposure measurement data, and frequency and likelihood of the worker's exposure. Preliminary evidence shows that for respiration filtration media there is no deviation from the classical single-fiber theory for particulates as small as 2.5 nm in diameter. While this evidence needs confirmation, NIOSH certified respirators will be useful for protecting workers from nanoparticles inhalation when properly selected and fit tested as part of a complete respiratory protection program. Use NIOSH approved positive flow mask if dust becomes airborne. Try to avoid creating dust conditions.

Eye protection: Use chemical safety goggles and/or full face shield where dusting or splashing of solution is possible. Maintain eye wash fountain in work area.

Skin Protection: Wear impervious protective clothing including boots, gloves, lab coat, apron or coveralls, as appropriate, to prevent skin contact. Wash thoroughly after handling. Maintain quick-drench facilities in work area.

Section 9: Physical and Chemical Properties



Appearance: gray suspension

Silver metal [7440-22-4]

Theoretical Density: 10.49 g/cm³
Bulk Density: 0.5 g/cm³
Molecular Weight: 107.868 AMU
pH: Not available
Boiling Point: 2212C (4014F)
Melting Point: 962C (1764F)
Vapor Density (Air=1): Not available
Vapor Pressure: Not available
Evaporation Rate: Not available
Viscosity: Not applicable
Decomposition Temp: Not available
Solubility in water: Not soluble

isopropanol [67-63-0]:

Odor: characteristic
Molecular Weight: 60.10
Vapor pressure @ 20C: 33 mm Hg
Vapor density: 2.1 (air =1)
Boiling point @ 760mm Hg: 82.3C (180F)
Freezing Point: -89C (-127F)
Solubility in Water: complete @ 20C
Specific Gravity @ 20C: .787
Evaporation Rate: 2.9 (butyl acetate = 1)
Percent Volatiles: 100%
Flash point: 12°C (Abel)
Explosion limit - upper: 12% (v/v)
Explosion limit - lower: 2% (v/v)
Auto-ignition temperature: 428°C
Other properties:

Section 10: Stability and Reactivity

Stability: Stable under ordinary conditions of use and storage. Reacts with strong oxidizing agents. Reacts with strong acids.

Conditions to avoid: Heat, flame, spark; Dust generation and incompatibles.

Incompatibility/Materials to avoid: strong oxidizing agents; strong inorganic acids; halogens; aldehydes; and halogen compounds. Silver is incompatible with acetylene, ammonia, strong hydrogen peroxide solutions, strong acids, oxalic acid, tartaric acid, bromoazide, chlorine trifluoride, and ethylenimine.

Hazardous Combustion/Decomposition Products: Carbon monoxide and/or carbon dioxide, Metal oxide fume.

Hazardous Polymerization: Will not occur

Section 11: Toxicological Information

Silver metal [7440-22-4]

NTP Known Carcinogen: No
NTP Anticipated Carcinogen: No
IARC Category: None



Isopropanol [87-83-0]:

Basis for assessment: Information given is based on product data.
Acute toxicity - oral: LD50 > 2000 mg/kg
Acute toxicity - dermal: LD50 > 2000 mg/kg
Acute toxicity - inhalation: LC50 > 5 mg/l
Eye irritation: Slight irritant.
Skin irritation: Slight irritant.
Respiratory irritation: Irritant in animal studies.
Skin sensitisation: May cause skin sensitisation.
(Sub) chronic toxicity: Repeated exposure causes liver damage.
Human effects: Repeated exposure can lead to allergic contact dermatitis. High exposures can cause drowsiness and dizziness. Can cause liver damage.

Section 12: Ecological Information**Silver metal [7440-22-4]**

Environmental Fate: No information found in our selected references.
Environmental Toxicity: No information found in our selected sources
Bioaccumulation: Not expected to occur.

Isopropanol [87-83-0]:**Environmental Fate:**

When released into the soil, this material is expected to quickly evaporate. When released into the soil, this material may leach into groundwater. When released into the soil, this material may biodegrade to a moderate extent. When released to water, this material is expected to quickly evaporate. When released into the water, this material is expected to have a half-life between 1 and 10 days. When released into water, this material may biodegrade to a moderate extent. This material is not expected to significantly bioaccumulate. When released into the air, this material is expected to be readily degraded by reaction with photochemically produced hydroxyl radicals. When released into the air, this material is expected to have a half-life between 1 and 10 days. When released into the air, this material may be removed from the atmosphere to a moderate extent by wet deposition.

Environmental Toxicity:

Bio-accumulation: Does not bio-accumulate.
Acute toxicity - fish: LC50 > 100 mg/l
Acute toxicity - daphnia: EC50 > 100 mg/l
Acute toxicity - algae: IC50 > 100 mg/l
Acute toxicity - bacteria: IC50 > 100 mg/l
Sewage treatment: Practically non-toxic, EC50 > 100 mg/l, to organisms in sewage treatment plants.
Other information: Poses a significant risk of oxygen depletion in aquatic systems.

Section 13: Disposal Considerations

This material and its container must be disposed of as hazardous waste.

Precautions: Refer to Section 7 before handling the products or containers.
Waste disposal: Recover or recycle if possible. Otherwise incineration.
Product disposal: Recover or recycle if possible. Otherwise incineration.
Container disposal: Drain container thoroughly. After draining, vent in a safe place away from sparks and fire. Vapors may collect in empty containers. Send to drum recoverer or metal reclaimmer. Residues may cause an explosion hazard. Do not puncture, cut, or weld uncleaned drums.
Processing, use, or contamination of this product may change the waste management options.



Dispose of spill-clean up and other wastes in accordance with Federal, State, and local regulations.

Section 14: Transport Information

Proper Shipping Name: Isopropanol
Hazard Class: 3
UN Number: 1219
Packaging Group II

IMO Information: Isopropanol
Label of Class: 3.2
Packaging Group II
Intermediate flashpoint group

Section 15: Regulatory Information

Ingredient	--Chemical Inventory Status - Part 1--			
	TSCA	EC	Japan	Australia
Isopropyl Alcohol (67-63-0)	Yes	Yes	Yes	Yes
Silver (7440-22-4)	Yes	not available-----		

Ingredient	--Chemical Inventory Status - Part 2--			
	Korea	DSL	NDSL	Phil.
Isopropyl Alcohol (67-63-0)	Yes	Yes	No	Yes
Silver (7440-22-4)	No	Yes	No	

Ingredient	--Federal, State & International Regulations - Part 1--			
	RQ	TPQ	List	-----SARA 313----- Chemical Catg.
Isopropyl Alcohol (67-63-0)	No	No	Yes	No
Silver (7440-22-4)			Yes	

Ingredient	--Federal, State & International Regulations - Part 2--		
	CERCLA	281.33	-----TSCA----- § (d)
Isopropyl Alcohol (67-63-0)	No	No	No
Silver (7440-22-4)	6000	No	No

Chemical Weapons Convention: No TSCA 12(b): No QDTA: Yes
SARA 311/312: Acute: Yes Chronic: Yes Fire: Yes Pressure: No Reactivity: No (Mixture / Liquid)
Australian Hazchem Code: 2[S]2
Poison Schedule: None allocated.

EC Label name:	Isopropanol
EC Classification:	Highly flammable
EC Symbols:	F, Xi
EC Safety phrases:	
S7	Keep container tightly closed
S16	Keep away from sources of ignition - no smoking.
R-phrases(s):	
R11:	Highly Flammable
R36:	Irritating to eyes
R67:	Vapors may cause drowsiness or



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dizziness
EINECS (EC): 200-661-7
EC Annex I Number: 603-003-00-0
MITI (Japan): 2-207
TSCA (USA): Listed
AICS (Australia): Listed
DSL (Canada): Listed

Section 16: Other Information

NFPA Ratings: Health: 1 Flammability: 3 Reactivity: 1

Label Hazard Warning:

WARNING! FLAMMABLE LIQUID AND VAPOR. HARMFUL IF SWALLOWED OR INHALED. CAUSES IRRITATION TO EYES AND RESPIRATORY TRACT. AFFECTS CENTRAL NERVOUS SYSTEM. MAY BE HARMFUL IF ABSORBED THROUGH SKIN. MAY CAUSE IRRITATION TO SKIN.

Label Precautions:

Keep away from heat, sparks and flame.
Keep container closed.
Use only with adequate ventilation.
Wash thoroughly after handling.
Avoid breathing vapor, mist, or dust.
Avoid contact with eyes, skin and clothing.

Label First Aid:

If swallowed, give large amounts of water to drink. Never give anything by mouth to an unconscious person. If inhaled, remove to fresh air. If not breathing, give artificial respiration. If breathing is difficult, give oxygen. In case of contact, immediately flush eyes or skin with plenty of water for at least 15 minutes. Remove contaminated clothing and shoes. Wash clothing before reuse. In all cases, get medical attention.

MSDS Creation Date: 2/06/2007

Last Updated: 08/01/09

The information above is believed to be accurate and represents the best information currently available to us and is applicable to the product with regard to appropriate safety precautions. It does not represent any guarantee of the properties of the product. This document is offered solely as a guide for the customer's consideration. Investigation and verification, and is intended only as a guide to the appropriate precautionary handling of the material by a properly trained person using this product. These suggestions do not replace state, municipal and/or insurance requirements. Any use of this information must be determined by the user to be in accordance with applicable Federal, state and local regulations. We make no warranty of merchantability or any other warranty, express or implied, with respect to such information, and we assume no liability resulting from its use. It is the user's responsibility to satisfy himself as to the suitability and completeness of such information for his own particular application. In no event shall Seashell Technology, LLC be liable for any claims, losses, or damages of any third party or for lost profits or any special, indirect, incidental, consequential or exemplary damages, howsoever arising, even if Seashell Technology, LLC has been advised of the possibility of such damages. See reverse side of invoice or packing slip for additional terms and conditions of sale.



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REFERENCES

References for Published Research on Nano Silver:

C. Levard, B. Reinsch, F.M. Michel, C. Oumahi, G. V. Lowry, and G. E. Brown, Jr., "Sulfidation Processes of PVP-Coated Silver Nanoparticles in Aqueous Solution: Impact on Dissolution Rate," *Environ. Sci. Technol.*, 2011, **45**, 5260-5266.

L. Hu, H. Wu, and Y. Cui, "Metal nanogrids, nanowires, and nanofibers for transparent electrodes," *MRS Bulletin*, 2011, **36**, 760-765.

W. M. Wang, R. M. Stoltenberg, S. Liu, and Z. Bao, "Direct Patterning of Gold Nanoparticles Using Dip-Pen Nanolithography," *ACS Nano*, 2008, **2**, 2135-2142.

Attachment 4:

Section D-F for Nano Titanium Dioxide

Section D: Nanomaterial Chemical and Physical Properties			
Product / Production Information (Based on three Stanford University academic research laboratories)			
Nano Chemical Name:		Nano Titanium Dioxide	
Commercial Name(s):		Ti-Nanoxide; Degussa P-25 TiO ₂ ; Titania	
Annual Production Volume:		N/A (purchased ~150 g)	
Production Method(s):		Unknown	
Identification of the Supplier(s):		Solaronix; Degussa; Dyesol	
Parameter		Value / Range ¹ (include units)	Name of Analytical Method(s) ²
Physical Properties			
Shape (Morphology)		Spherical	From Suppliers
Density		4.23 g/cm ³	From Suppliers
Surface Area		50-80 m ² /g	From Suppliers
Particle Size Distribution	Air	15-18 nm	From Suppliers
	Liquid	15-20 nm	Transmission Electron Microscopy
	Solid / Powder	15-80 nm	Transmission Electron Microscopy, Scanning Electron Microscopy
Other (Specify)			
Chemical Properties			
Chemical Composition		TiO ₂	X-Ray Diffraction
Surface Modification (Coating, Functionalization)		Unknown	Unknown
Purity		~3% - 99%	From Suppliers
Surface Charge		Unknown	Unknown
Dispersion ³	Air	N/A (paste); Unknown (powder)	N/A (paste); Unknown (powder)
	Liquid	Disperses in Ethanol (paste); Unknown (powder)	Visible examination (paste); Unknown (powder)
	Solid	N/A (paste); Unknown (powder)	N/A (paste); Unknown (powder)
Identifying and Determining Concentration of Nano Chemical, Its Metabolites, and Degradation Products in Specified Matrices Water, Air, Soil, Sediment, Sludge, Chemical Waste, Fish, Blood, Adipose Tissue, Urine, Other (Specify)		See Section F	
Solubility	Water Solubility	None (paste); Unknown (powder)	Visible examination (paste); Unknown (powder)
	Solubility in Organic Solvent	Slightly soluble in ethanol (paste); Unknown (powder)	Visible examination (paste); Unknown (powder)
n-Octanol- Water Partition Coefficient		Unknown	Unknown

Stability and Reactivity	Flammability	None (paste); Unknown (powder)	From Supplier (paste); Unknown (powder)
	Explosiveness	None (paste); Unknown (powder)	From Supplier (paste); Unknown (powder)
	Oxidizing Properties	None (paste); Unknown (powder)	From Supplier (paste); Unknown (powder)
	Oxidation Reduction Potential	-0.25 V vs. SCE (paste); Unknown (powder)	From Scientific Journal (paste); Unknown (powder)
Stability and Reactivity	Storage Stability and Reactivity (Container Material)	Stable (paste); Unknown (powder)	From Supplier (paste); Unknown (powder)
	Stability to Thermal, Sunlight, and Metal(s)	Stable, plastic container (paste); Unknown (powder)	From Supplier (paste); Unknown (powder)

Section E: Globally Harmonized System Safety Data Sheet (SDS) or Materials Safety Data Sheet (MSDS)

<i>Chemical Name</i>	<i>Source</i>
Nano Titanium Dioxide	Solaranix; Degussa; Dyesol

Section F: Describe the analytical test method(s) that you use or plan to use to sample, prepare and analyze a specific matrix (water, air, soil, sediment, sludge, chemical waste, fish, blood, adipose tissue, and urine) to determine the identity and concentration of each specified nanomaterial.

<i>Nanomaterial</i>	<i>Purpose (sample, prepare, analyze)</i>	<i>Matrix</i>
Nano Titanium Dioxide	N/A	N/A
Method description		
N/A		

MATERIAL SAFETY DATA SHEETS

Safety data sheet

according to 1907/2006/EC, Article 31

Printing date 01.05.2011

Version number 1

Revision: 01.05.2011

1 Identification of the substance/mixture and of the company/undertaking

- Product identifier
- Trade name: **Ti-Nanoxide T-L**
- Relevant identified uses of the substance or mixture and uses advised against
- Application of the substance / the preparation Transparent Nanocrystalline Titanium Dioxide
- Details of the supplier of the safety data sheet
- Manufacturer/Supplier:
SOLARONIX SA
Rue de l'Ouriette 129, CH-1170 Aubonne, Switzerland
info@solaronix.com
T +41 21 821 22 80
www.solaronix.com
F +41 21 821 22 89
info@solaronix.com
- Emergency telephone number: Swiss Toxicological Information Center: +41 44 251 51 51

2 Hazards identification

- Classification of the substance or mixture
- Classification according to Regulation (EC) No 1272/2008

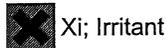


GHS07

Skin Irrit. 2 H315 Causes skin irritation.

Eye Irrit. 2 H319 Causes serious eye irritation.

- Classification according to Directive 67/548/EEC or Directive 1999/45/EC



Xi; Irritant

R36/38: Irritating to eyes and skin.

- Information concerning particular hazards for human and environment:
The product has to be labelled in the latest valid version according to the calculation procedure of the "General Classification guideline for preparations of the EU" .

- Classification system:

The classification is according to the latest editions of the EU-lists, and extended by company and literature data.

- Label elements

- Labelling according to Regulation (EC) No 1272/2008

The product is classified and labelled according to the CLP regulation.

- Hazard pictograms



GHS07

- Signal word Warning

- Hazard statements

H315 Causes skin irritation.

H319 Causes serious eye irritation.

- Precautionary statements

P280

Wear protective gloves/protective clothing/eye protection/face protection.

(Contd. on page 2)

Safety data sheet

according to 1907/2006/EC, Article 31

Printing date 01.05.2011

Version number 1

Revision: 01.05.2011

 Trade name: **Ti-Nanoxide T-L**

(Contd. of page 1)

- P305+P351+P338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
- P321 Specific treatment (see on this label).
- P362 Take off contaminated clothing and wash before reuse.
- P332+P313 If skin irritation occurs: Get medical advice/attention.
- P337+P313 If eye irritation persists: Get medical advice/attention.

- **Other hazards**

- **Results of PBT and vPvB assessment**

- PBT: Not applicable.

- vPvB: Not applicable.

3 Composition/information on ingredients

- **Chemical characterization: Mixtures**

- **Description:**

Paste containing about 11% wt. of 15-20 nm titanium dioxide (TiO₂) anatase particles CAS 1317-70-0

- **Dangerous components:**

78989-43-2	Red fuming nitric acid	≤2.5%
	 C R35;  O R8	
	 Ox. Liq. 2, H272;  Skin Corr. 1A, H314	

- **Additional information:** For the wording of the listed risk phrases, refer to section 16.

4 First aid measures

- **Description of first aid measures**

- **After excessive inhalation:**

In case of unconsciousness place patient in a stable laying down side position for transportation.

- **After skin contact:** Immediately wash with water and soap and rinse thoroughly.

- **After eye contact:**

Rinse opened eye for several minutes under running water. If symptoms persist, consult a doctor.

- **After swallowing:** If symptoms persist consult a doctor.

- **Information for doctor:**

- **Most important symptoms and effects, both acute and delayed**

No further relevant information available.

- **Indication of any immediate medical attention and special treatment needed**

No further relevant information available.

5 Firefighting measures

- **Suitable extinguishing agents:**

CO₂, powder or water spray. Fight larger fires with water spray or alcohol resistant foam.

- **Special hazards arising from the substance or mixture**

No further relevant information available.

- **Advice for firefighters**

- **Protective equipment:** No special measures required.

CB

(Contd. on page 3)

Safety data sheet

according to 1907/2006/EC, Article 31

Printing date 01.05.2011

Version number 1

Revision: 01.05.2011

Trade name: **Ti-Nanoxide T-L**

(Contd. of page 2)

6 Accidental release measures

- **Personal precautions, protective equipment and emergency procedures** Not required.
- **Environmental precautions:**
 - Dilute with plenty of water.
 - Do not allow to enter sewers/ surface or ground water.
- **Methods and material for containment and cleaning up:**
 - Absorb with liquid-binding material (sand, diatomite, acid binders, universal binders, sawdust).
- **Reference to other sections**
 - See Section 7 for information on safe handling.
 - See Section 8 for information on personal protective equipment.
 - See Section 13 for disposal information.

7 Handling and storage

- **Handling:**
- **Precautions for safe handling** No special precautions are necessary if used correctly.
- **Information about fire - and explosion protection:** No special measures required.
- **Conditions for safe storage, including any incompatibilities**
- **Storage:**
- **Requirements to be met by storerooms and receptacles:** No special requirements.
- **Information about storage in one common storage facility:** Not required.
- **Further information about storage conditions:** Keep container tightly sealed.
- **Specific end use(s)** No further relevant information available.

8 Exposure controls/personal protection

- **Additional information about design of technical facilities:** No further data; see section 7.
- **Control parameters**
- **Ingredients with limit values that require monitoring at the workplace:**
 - The product does not contain any significant quantities of materials with critical values that have to be monitored at the workplace.
- **Additional information:**
 - As a basis for the production of this document, the most current valid lists were used.
- **Exposure controls**
- **Personal protective equipment:**
- **General protective and hygienic measures:** Keep away from food, beverages and petfood. Immediately remove all soiled and contaminated clothing Wash hands before breaks and at the end of work. Avoid contact with the eyes and skin.
- **Respiratory protection:** Not required.
- **Protection of hands:**



Protective gloves

The glove material has to be impermeable and resistant to the product/ the substance/ the preparation.

(Contd. on page 4)

CB

Safety data sheet
according to 1907/2006/EC, Article 31

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Trade name: Ti-Nanoxide T-L

(Contd. of page 3)

· Gloves material :

The selection of suitable gloves does not only depend on the material, but also on further marks of quality which may vary from manufacturer to manufacturer. As the product is a preparation of several substances, the resistance of the glove material can not be calculated in advance and has therefore to be checked before use.

· Eye protection:


Tightly sealed goggles

9 Physical and chemical properties

· Information on basic physical and chemical properties
· General Information
· Appearance:

Form: Liquid
Colour: According to product specification

· Odour: Characteristic

· Odour threshold: Not determined.

· pH-value: Not determined.

· Change in condition

Melting point/Melting range: Undetermined.
Boiling point/Boiling range: 100°C

· Flash point: Not applicable.

· Flammability (solid, gaseous): Not applicable.

· Ignition temperature: Not applicable.

· Decomposition temperature: Not determined.

· Self-igniting: Product is not self igniting.

· Danger of explosion: Product does not present an explosion hazard.

· Explosion limits:

Lower: Not determined.
Upper: Not determined.

· Vapour pressure at 20°C: 23 hPa

· Density: Not determined.

· Relative density: Not determined.

· Vapour density: Not determined.

· Evaporation rate: Not determined.

· Solubility in / Miscibility with

water: Fully miscible.

· Segregation coefficient (n-octanol/water): Not determined.

· Viscosity:

Dynamic: Not determined.

(Contd. on page 5)

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according to 1907/2006/EC, Article 31

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Trade name: **Ti-Nanoxide T-L**

(Contd. of page 4)

Kinematic:	Not determined.
· Other information	No further relevant information available.

10 Stability and reactivity

- **Reactivity**
- **Chemical stability**
- **Thermal decomposition / conditions to be avoided:**
No decomposition if used according to specifications.
- **Possibility of hazardous reactions** No dangerous reactions known.
- **Conditions to avoid** No further relevant information available.
- **Incompatible materials:** No further relevant information available.
- **Hazardous decomposition products:** No dangerous decomposition products known.

11 Toxicological information

- **Information on toxicological effects**
- **Acute toxicity:**
- **Primary irritant effect:**
- **On the skin:** Irritant to skin and mucous membranes.
- **On the eye:** Irritating effect.
- **Sensitization:** No sensitizing effects known.
- **Additional toxicological information:**
The product shows the following hazards according to the calculation method of the General EU Classification Guidelines for Preparations as issued in the latest version:
Irritant

12 Ecological information

- **Toxicity**
- **Aquatic toxicity:** No further relevant information available.
- **Persistence and degradability** No further relevant information available.
- **Behaviour in environmental systems:**
- **Bioaccumulative potential** No further relevant information available.
- **Mobility in soil** No further relevant information available.
- **Additional ecological information:**
- **General notes:**
Water hazard class 1 (German Regulation) (Self-assessment): slightly hazardous for water.
Do not allow undiluted product or large quantities of it to reach ground water, water course or sewage system.
- **Results of PBT and vPvB assessment**
- **PBT:** Not applicable.
- **vPvB:** Not applicable.
- **Other adverse effects** No further relevant information available.

(Contd. on page 6)

Safety data sheet
according to 1907/2006/EC, Article 31

Printing date 01.05.2011

Version number 1

Revision: 01.05.2011

Trade name: Ti-Nanoxide T-L

(Contd. of page 5)

13 Disposal considerations

- Waste treatment methods
- Recommendation



Must not be disposed of together with household garbage. Do not allow product to reach sewage system.

- **Uncleaned packaging:**
- **Recommendation:** Disposal must be made according to official regulations.
- **Recommended cleansing agents:** Water, if necessary together with cleansing agents.

14 Transport information

- Land transport ADR/RID (cross-border) :
- ADR/RID class: -

- Maritime transport IMDG:
- IMDG Class: -
- Marine pollutant: No

- Air transport ICAO-TI and IATA-DGR:
- ICAO/IATA Class: -

- UN "Model Regulation": -
- Special precautions for user Not applicable.
- Transport in bulk according to Annex II of MARPOL73/78 and the IBC Code Not applicable.

15 Regulatory information

- **Chemical safety assessment:** A Chemical Safety Assessment has not been carried out.

16 Other information

This information is based on our present knowledge. However, this shall not constitute a guarantee for any specific product features and shall not establish a legally valid contractual relationship.

- **Relevant phrases**

H272 May intensify fire; oxidiser.

H314 Causes severe skin burns and eye damage.

R35 Causes severe burns.

R8 Contact with combustible material may cause fire.

- **Abbreviations and acronyms:**

ADR: Accord européen sur le transport des marchandises dangereuses par Route (European Agreement concerning the International Carriage of Dangerous Goods by Road)

RID: Règlement international concernant le transport des marchandises dangereuses par chemin de fer (Regulations Concerning the International Transport of Dangerous Goods by Rail)

IMDG: International Maritime Code for Dangerous Goods

IATA: International Air Transport Association

IATA-DGR: Dangerous Goods Regulations by the "International Air Transport Association" (IATA)

ICAO: International Civil Aviation Organization

ICAO-TI: Technical Instructions by the "International Civil Aviation Organization" (ICAO)

GHS: Globally Harmonized System of Classification and Labelling of Chemicals

REFERENCES

Reference for Published Research on Nano Titanium Dioxide:

I. S. Cho, Z. Chen, A. J. Forman, D. R. Kim, P. M. Rao, T. F. Jaramillo, and X. Zheng,
"Branched TiO₂ Nanorods for Photoelectrochemical Hydrogen Production," *Nano Lett.*,
2011, **11**, 4978-4984.



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Low Temperature Titania

- Ti-Nanoxide D-L
- Ti-Nanoxide T-L
- Ti-Nanoxide HT-L

[Related Products](#)

Versatile Titania

- Ti-Nanoxide D
- Ti-Nanoxide T
- Ti-Nanoxide HT

[Comparison chart](#)

Ti-Nanoxide T-L

Low Temperature Transparent Nanocrystalline Titanium Dioxide

Ti-Nanoxide T-L is an acidic colloidal paste in a mostly alcoholic vehicle. It contains about 11 % wt. nanocrystalline titanium dioxide. This paste is well suited for coating heat sensitive substrates, such as polymers.

Recommended for doctor-blade (squeegee) printing. The sintering of this paste can be done starting at 120 °C and the obtained layer is transparent.



Buy Now

Conditioning:

- 10 g
- 20 g
- 50 g
- 100 g
- 200 g
- 500 g
- 1000 g

Please feel free to contact us for larger quantities or repetitive orders of this product.

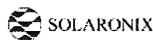
[Contact us](#)

Customer adapted formulations of this product are available upon request.

Specifications

<i>Product designation:</i>	Ti-Nanoxide T-L
<i>Anatase particles:</i>	15-20 nm
<i>Concentration:</i>	~11 % wt.
<i>Diffusing particles:</i>	none
<i>Vehicle:</i>	ethanol, water, acids

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SAFETY DATA SHEET (EC 1907/2006)**AEROXIDE® TiO2 P 25**

Material no.		Version	3.8 / REG_EU
Specification	132843	Revision date	29.08.2008
VA-Nr		Print Date	30.08.2008
		Page	1 / 6

1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND OF THE COMPANY/UNDERTAKING**Product information**

Trade name : AEROXIDE® TiO2 P 25

Company : Evonik Degussa GmbH
Inorganic Materials
Produktsicherheit IM-IM-PS
Postfach 1345
D-63403 Hanau

Telephone : +49 (0)6181 59-4787
Telefax : +49 (0)6181 59-4205
Email address : sds.asfp@evonik.com
Emergency telephone number : +49 (0)7623-919191

Use of the Substance /
Preparation : Catalyst support
Stabilizer
UV-filters

2. HAZARDS IDENTIFICATION**Additional safety information for humans and the environment**

On the basis of our data the product is not a hazardous substance as defined by the Chemicals Act or Hazardous Substance Ordinance in the currently valid versions.

3. COMPOSITION/INFORMATION ON INGREDIENTS**Information on ingredients / Hazardous components**

• **Titanium dioxide**
CAS-No. 13463-67-7 EC-No. 236-675-5

See chapter 16 for text of risk phrases

4. FIRST AID MEASURES**Inhalation**

In case product dust is released:
Possible discomfort: cough, sneezing
Move victims into fresh air.

Skin contact

Wash off with plenty of water and soap.

Eye contact

Possible discomfort is due to foreign substance effect.
Rinse thoroughly with plenty of water keeping eyelid open.
In case of persistent discomfort: Consult an ophthalmologist.

SAFETY DATA SHEET (EC 1907/2006)**AEROXIDE® TiO2 P 25**

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Ingestion

Clean mouth with water and drink afterwards plenty of water.
After absorbing large amounts of substance / In case of discomfort: Supply with medical care.

Notes to physician

No hazards which require special first aid measures.

5. FIRE-FIGHTING MEASURES**Suitable extinguishing media**

All extinguishing substances suitable.

Specific hazards during fire fighting

None known

Further information

Water used to extinguish fire should not enter drainage systems, soil or stretches of water.
Ensure there are sufficient retaining facilities for water used to extinguish fire.
Fire residues and contaminated fire extinguishing water must be disposed of in accordance with local regulations.

6. ACCIDENTAL RELEASE MEASURES**Personal precautions**

Wear personal protective equipment.

Environmental precautions

Do not allow entrance in sewage water, soil stretches of water, groundwater, drainage systems.

Methods for cleaning up

Sweep up or vacuum up spillage and collect in suitable container for disposal.
Avoid dust formation.

7. HANDLING AND STORAGE**Handling****Safe handling advice**

If necessary: Local ventilation.

Advice on protection against fire and explosion

Take precautionary measures against static discharges.

Storage**Requirements for storage areas and containers**

Keep in a dry place.

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8. EXPOSURE CONTROLS / PERSONAL PROTECTION**Components with workplace control parameters****Personal protective equipment****Respiratory protection**

No special protective equipment required.
If dust occurs: Dust mask with P2 particle filter

Hand protection

Wear protective gloves made of the following materials: nitrile rubber (NBR), butyl rubber, PVC.
The material thickness and rupture time data do not apply to non-solute solids / dusts.

Eye protection

Safety glasses with side-shields
If dust occurs: basket-shaped glasses

Skin and body protection

No special protective equipment required.
preventive skin protection
Cleanse and apply cream to skin after work.

Hygiene measures

When using, do not eat, drink or smoke. Wash face and/or hands before break and end of work.
Avoid contaminating clothes with product. Wash contaminated clothing after use.

Protective measures

Handle in accordance with good industrial hygiene and safety practices.
If there is the possibility of skin/eye contact, the indicated hand/eye/body protection should be used.
If the limits at the workplace are exceeded and/or larger amounts are released (leakage, spilling, dust) the indicated respiratory protection should be used.

9. PHYSICAL AND CHEMICAL PROPERTIES**Appearance**

Form	powder
Colour	white
Odour	odourless

Safety data

pH	3.5 - 4.5	(40 g / l)	(20 °C)
Melting point/range	ca. 1850 °C		
Boiling point/range	not applicable		
Flash point	not applicable		
Flammability	not applicable		
Ignition temperature	not applicable		
Autoinflammability	not applicable		

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Lower explosion limit	not applicable
Upper explosion limit	not applicable
Minimum ignition energy	> 10 Joule
Vapour pressure	not applicable
Density	ca. 3.8 g/cm ³ (20 °C)
Tapped density	ca. 130 g / l Method: DIN 53 194
Water solubility	insoluble
Partition coefficient (n-octanol/water)	not applicable
Viscosity, dynamic	not applicable

10. STABILITY AND REACTIVITY

Hazardous decomposition products	None known
Thermal decomposition	> 2000 °C

11. TOXICOLOGICAL INFORMATION

Acute oral toxicity	LD50 Rat: > 10000 mg/kg Method: literature (limit test)
Acute dermal toxicity	LD50 Rabbit: >= 10000 mg/kg Method: literature
Skin irritation	Rabbit / literature not irritating
Eye irritation	Rabbit / literature not irritating
Sensitization	Optimizations-test guinea pig: not sensitizing Method: literature Patch test : not sensitizing Method: literature
Gentoxicity in vitro	Microorganisms, cell cultures Shown no mutagenic/genotoxic effect., literature
Gentoxicity in vivo	Microorganisms, cell cultures Shown no mutagenic/genotoxic effect., literature
Carcinogenicity	Oral rat, mouse: 103 weeks

SAFETY DATA SHEET (EC 1907/2006)**AEROXIDE® TiO2 P 25**

Material no.		Version	3.8 / REG_EU
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no evidence that cancer may be caused, literature.

Feeding experiments

inhalative Rat: 2 years

Method: literature

Increased incidence of lung tumors.

The scientific discussion of the tumorigenic effect of sparingly soluble inorganic particles (fine dusts)- such as titanium dioxide - is ongoing. It is the opinion of many inhalation toxicologists that the tumor formation observed in rats results from a species-specific mechanism involving overloading of the rat lung (overload phenomenon). Corresponding findings resulting from exposure of humans have not been observed to date. On the other hand, the International Agency for Research on Cancer (IARC) assessed, in February of 2006, the available rat model studies as constituting sufficient proof of the carcinogenicity of titanium dioxide in animal models. For humans, the IARC does not see sufficient evidence of a carcinogenic effect of titanium dioxide. However, the IARC evaluation scheme results in an overall assessment of titanium dioxide as "possibly carcinogenic to humans" (Group 2B).

inhalative (mouse): 2 years

no evidence that cancer may be caused, literature.

Human experience

Epidemiological studies to date have not revealed any evidence of a relation between exposure to titanium dioxide and diseases of the respiratory tract beyond general effects of dust.

12. ECOLOGICAL INFORMATION**Elimination information (persistence and degradability)****Behaviour in environmental compartments****Ecotoxicity effects**

Toxicity to fish	LC50 Fundulus heteroclitus: > 1000 mg/l / 96 h Method: literature
Toxicity to daphnia	EC0 Daphnia magna: 1000 mg/l / 48 h Method: literature
Toxicity to bacteria	EC0 Pseudomonas fluorescens: 10000 mg/l / 24 h Method: DEV, DIN 38412, T. 8 (modified).

13. DISPOSAL CONSIDERATIONS**Product**

Disposal according to local authority regulations.

Uncleaned packaging

Offer rinsed packaging material to local recycling facilities.

Other countries: observe the national regulations.

SAFETY DATA SHEET (EC 1907/2006)**AEROXIDE® TiO2 P 25**

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Waste Key Number

No waste key number as per the European Waste Types List can be assigned to this product, since such classification is based on the (as yet undetermined) use to which the product is put by the consumer.

The waste key number must be determined as per the European Waste Types List (decision on EU Waste Types List 2000/532/EC) in cooperation with the disposal firm / producing firm / official authority.

14. TRANSPORT INFORMATION**Transport/further information**

Not classified as dangerous in the meaning of transport regulations.

15. REGULATORY INFORMATION**Labelling according to EC Directives**

Other data

On the basis of our data the product is not a hazardous substance as defined by the Chemicals Act or Hazardous Substance Ordinance in the currently valid versions.

National legislation

16. OTHER INFORMATION**Risk phrase (R phrase) texts****Further information**

Changes since the last version are highlighted in the margin. This version replaces all previous versions.

The information provided in this Safety Data Sheet is correct to the best of our knowledge, information and belief at the date of its publication. The information given is designed only as a guidance for safe handling, use, processing, storage, transportation, disposal and release and is not to be considered a warranty or quality specification. The information relates only to the specific material designated and may not be valid for such material used in combination with any other materials or in any process, unless specified in the text.

MATERIAL SAFETY DATA SHEET

SECTION 1 IDENTIFICATION OF THE MATERIAL AND SUPPLIER

PRODUCT NAME: DSL 18NR-T (Transparent)

PRODUCT DESCRIPTION: Paste with titanium dioxide particles approximately 20nm in diameter.

USE: Screen printing of working electrodes for Dye Solar Cells and other devices utilising nanoporous titanium dioxide.

MANUFACTURER:

Company: Dyesol Australia Pty Ltd
Address: 3 Dominion Place, Queanbeyan
New South Wales 2620
Australia
Telephone: (+61) (2) 6299 1592
Fax: (+61) (2) 6299 1698
Email: information@dyesol.com
Emergency: (+61) (2) 6299 1592

SECTION 2 HAZARDS IDENTIFICATION

HAZARD CLASSIFICATION: HAZARDOUS SUBSTANCE. NON-DANGEROUS GOODS. According to the Criteria of NOHSC.

RISK PHRASE(S): Irritant (Xi) R36/38

SAFETY PHRASE(S): S24: Avoid contact with skin
S25: Avoid contact with eyes
S26: In case of contact with eyes, rinse immediately with plenty of water and seek medical advice

SECTION 3 COMPOSITION/INFORMATION ON INGREDIENTS**INGREDIENTS**

NAME	CAS RN	PROPORTION
		wt%
Titanium Dioxide	1317-70-0	10 - 30 %
Ethyl cellulose	9004-57-3	5 - 15 %
Terpineol	8006-39-1 8001-47-7	50 - 70%
Organic plasticiser		5 - 20%

SECTION 4 FIRST AID MEASURES

If poisoning occurs, contact a doctor or Poisons Information Centre.

In Australia phone 13 11 26

AFTER INHALATION

If excessive fumes or combustion products are inhaled, remove to fresh air, lay patient down and ensure clear breathing passages. In case of irritation or discomfort seek immediate medical advice.

AFTER SKIN CONTACT

The material may be mildly discomforting to the skin.
Open cuts, abraded or irritated skin should not be exposed to this material.
The material may accentuate a pre-existing skin condition.

AFTER EYE CONTACT

In case of contact with eyes, flush with copious amounts of water for at least 15 minutes. Assure adequate flushing by separating eyelids with finger. Seek medical advice.

IF SWALLOWED

Considered an unlikely route of entry in commercial/industrial/scientific environment
The material should be considered an irritant and may be harmful if swallowed in large quantity.

SECTION 5 FIRE FIGHTING MEASURES

FIRE FIGHTING MEASURES

The fire fighting measures are those applicable to the most flammable component, terpineol.

If safe, remove containers from path of fire.

Wear self-contained respirator and fully protective impervious suit.

SUITABLE EXTINGUISHING MEDIA

Foam, dry chemical powder, carbon dioxide, or water spray (large fires only).

HAZARDS FROM COMBUSTION PRODUCTS

In case of fire toxic fumes of Carbon Oxides (CO_x) can be released.

PRECAUTIONS FOR FIRE FIGHTERS AND SPECIAL PROTECTIVE EQUIPMENT

Wear self-contained respirator and fully protective impervious suit.

SECTION 6 ACCIDENTAL RELEASE MEASURES

EMERGENCY PROCEDURES

Wear respirator, chemical safety goggles, rubber boots, and heavy rubber gloves.

SPILLS, LEAKS OR RELEASES

Clean up all spills immediately.

Avoid contact with skin and eyes (gloves and safety glasses).

Use dry clean up procedures and avoid generating dust.

Ventilate area and wash spill site after material pickup is complete.

SECTION 7 HANDLING AND STORAGE

PRECAUTIONS FOR SAFE HANDLING

Avoid contact with eyes, skin, and clothing.

Do not eat and/or drink whilst using this material.

Do not eat, drink, or smoke in contaminated areas.

Wash hands thoroughly before eating.

Remove contaminated clothing and protective equipment before entering general or eating areas.

CONDITIONS FOR SAFE STORAGE

Keep away from sources of ignition.

Keep container tightly sealed and store in a cool, dark, dry, well ventilated area.

Avoid strong oxidising agents, strong bases.

No special precautions are necessary if used correctly.

Decomposition will not occur if used according to specifications.

No dangerous reactions are known.

SUITABLE CONTAINERS

Glass or plastic is suitable. No special precautions are required.

SECTION 8 EXPOSURE CONTROLS/PERSONAL PROTECTION

Not fully tested

PERSONAL PROTECTIVE EQUIPMENT

EYE

Wear safety glasses.

HANDS

Wear general impervious protective gloves, eg. disposable light weight plastic gloves.

OTHER

Impervious protective work clothing.

Eyewash unit. Properly operating chemical fume hood

Observe the general safety regulations when handling chemicals.

SECTION 9 PHYSICAL AND CHEMICAL PROPERTIES**PHYSICAL DESCRIPTION/PROPERTIES****APPEARANCE**

Orange/yellow viscous paste

PROPERTIES

The following is based on the properties of the solvent terpineol, being the predominant component.

Boiling Point (degC):	214-224
Melting Point (degC):	0-2
Vapour Pressure (kPa):	1.4 @ 20 C
Specific Gravity:	0.93-0.94 (~ 1.4 g/cc for product)
Flammability	Slight fire hazard when exposed to heat or flame.
Flash Point (degC):	90
Danger of explosion:	Heating may cause expansion or decomposition leading to violent rupture of containers.
Upper Explosive Limit (%)	Not available
Lower Explosive Limit (%)	Not available
Solubility in water:	Partly miscible

SECTION 10 STABILITY AND REACTIVITY

Decomposition will not occur if used according to specifications.

No dangerous reactions are known.

No special precautions are necessary if used correctly.

CONDITIONS TO AVOID

Keep container tightly sealed and store below 50°C.

INCOMPATIBLE MATERIALS

Avoid mixing with strong oxidisers

HAZARDOUS DECOMPOSITION PRODUCTS

In the event of a fire; see section 5

HAZARDOUS REACTIONS

None known.

SECTION 11 TOXICOLOGICAL INFORMATION

Product not fully tested

ACUTE HEALTH EFFECTS**SWALLOWED**

Considered an unlikely route of entry in commercial/industrial/scientific environment
The material should be considered irritant and may be harmful if swallowed in large quantity.

If swallowed, terpeneols can cause stomach inflammation with internal bleeding, heartburn, vomiting, diarrhoea, confusion, inco-ordination, a general malaise, headache, weakness, decreased body temperature, excitement, drowsiness, vertigo, convulsion, other central nervous effects or respiratory depression.

EYE

The material may be moderately discomforting to the eyes and is capable of causing temporary redness, temporary impairment of vision and/or other transient eye damage.

SKIN

The material may be mildly discomforting to the skin.

Open cuts, abraded or irritated skin should not be exposed to this material.

The material may accentuate a pre-existing skin condition.

INHALED

The vapour from the terpeneol in the paste may be discomforting to the upper respiratory tract, especially at higher temperatures.

CHRONIC HEALTH EFFECTS

Principal routes of exposure are usually by skin contact/absorption and inhalation of generated dust.

No human exposure data is available. For this reason health effects described are based on experience with chemically related materials.

As with any chemical product, contact with unprotected bare skin; inhalation of vapour, mist or dust in work place atmosphere; or ingestion in any form, should be avoided by observing good occupational work practice.

To the best of our knowledge the acute and chronic toxicity of this substance is not fully known.

ADDITIONAL TOXICOLOGICAL INFORMATION

None available

SECTION 12 ECOLOGICAL INFORMATION

No specific quantitative data available for this product.

SECTION 13 DISPOSAL CONSIDERATIONS

Consult state, local or national regulations to ensure proper disposal.
Disposal has to be carried out according to official regulations.

SECTION 14 TRANSPORT INFORMATION

Not a hazardous material for transportation.

SECTION 15 REGULATORY INFORMATION**INDICATION OF DANGER**

RISK PHRASE(S): Irritant (Xi) R36/38: Irritating to eyes and skin

SAFETY PHRASE(S): S24: Avoid contact with skin

S25: Avoid contact with eyes

S26: In case of contact with eyes, rinse immediately with
plenty of water and seek medical advice

SECTION 16 OTHER INFORMATION

Date of Issue: -29-July-2008

Employers should make independent judgement of suitability of this information to ensure proper use and protect health and safety of employees. The information in this Material Safety Data Sheet is furnished without any warranty. Any use of the product not in conformance with the Material Safety Data Sheet, or in combination with any other product or process, is the responsibility of the user.

For research and development, not for drug, household or other uses.

References:

- *National Code of Practice for the Preparation of Material Safety Data Sheets 2nd Edition*, [NOHSC:2011(2003)]
- *Approved Criteria For Classifying Hazardous Substances*, [NOHSC:1008(2004)]
- *Australian Dangerous Goods Code 6th edition*, Vol 1 incorporating corrigendum, National Road Transport Commission (Australia), Australian Department of Transport and Regional Services
- *Hazardous Substances Information System*, Australian Government - Department of Employment and Workplace relations – Office of the Australian Safety and Compensation Council <http://www.nohsc.gov.au/applications/hsis/>
- Chemwatch Material Safety Data Sheet for Titanium Dioxide, Ethyl cellulose and Terpineol

Attachment 5:

Section D-F for Nano Zinc Oxide

Section D: Nanomaterial Chemical and Physical Properties			
Product / Production Information (Based on two Stanford University academic research laboratories)			
Nano Chemical Name:		Nano Zinc Oxide	
Commercial Name(s):		N/A	
Annual Production Volume:		11 g	
Production Method(s):		Precipitation in Methanol at 60 °C, Hydrothermal synthesis	
Identification of the Supplier(s):		N/A (synthesized and used by SU academic research laboratory)	
Parameter		Value / Range ¹ (include units)	Name of Analytical Method(s) ²
Physical Properties			
Shape (Morphology)		Spheres, Nanowires	Scanning Electron Microscopy, Transmission Electron Microscopy
Density		5.6 g/cm ³	Theoretical
Surface Area		Unknown	Unknown
Particle Size Distribution	Air	Unknown	Unknown
	Liquid	4 nm; 30-100 nm diameter x 300-2000 nm length	Scanning Electron Microscopy, Transmission Electron Microscopy
	Solid / Powder	4 nm (spheres) Unknown (nanowires)	Transmission Electron Microscopy Unknown
Other (Specify)			
Chemical Properties			
Chemical Composition		ZnO	Inductively Coupled Plasma-Optical Emission Spectrometry, Transmission Electron Microscopy, X-ray Diffraction
Surface Modification (Coating, Functionalization)		Acetate	X-Ray Photoelectron Spectroscopy
Purity		Unknown	Unknown
Surface Charge		To Be Developed	To Be Developed
Dispersion ³	Air	Unknown	Unknown
	Liquid	Aggregated	Dynamic Light Scattering
	Solid	Aggregated	Dynamic Light Scattering
Identifying and Determining Concentration of Nano Chemical, Its Metabolites, and Degradation Products in Specified Matrices Water, Air, Soil, Sediment, Sludge, Chemical Waste, Fish, Blood, Adipose Tissue, Urine, Other (Specify)		See Section F	
Solubility	Water Solubility	10-50% (spheres) Unknown (Nanowires)	Inductively Coupled Plasma-Optical Emission Spectrometry Unknown
	Solubility in Organic Solvent	Unknown	Unknown

n-Octanol- Water Coefficient	Unknown	Unknown	
Stability and Reactivity	Flammability	Not Flammable	Based on bulk ZnO
	Explosiveness	Not Explosive	Based on bulk ZnO
	Oxidizing Properties	No	Based on bulk ZnO
	Oxidation Reduction Potential	No	Based on bulk ZnO
	Storage Stability and Reactivity (Container Material)	Unknown (spheres); stable (nanowires)	Unknown (spheres); visible observation (nanowires)
	Stability to Thermal, Sunlight, and Metal(s)	Production of Reactive Oxygen Species (spheres); Stable (nanowires)	Described in literature(spheres); Visible Observation (wires)

Section E: Globally Harmonized System Safety Data Sheet (SDS) or Materials Safety Data Sheet (MSDS)

Chemical Name	Source
Zinc Oxide nanoparticles, Zinc Oxide nanowires	None developed

Section F: Describe the analytical test method(s) that you use or plan to use to sample, prepare and analyze a specific matrix (water, air, soil, sediment, sludge, chemical waste, fish, blood, adipose tissue, and urine) to determine the identity and concentration of each specified nanomaterial.

Nanomaterial	Purpose (sample, prepare, analyze)	Matrix
Zinc Oxide nanoparticles	Analyze	Water
Method description		
Measure the solubility of Zinc Oxide solutions ranging from 1 ppb to 1000 ppm and its environmental transformations (i.e., reactions with Sulfur) utilizing Inductively Coupled Plasma-Optical Emission Spectrometry.		