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## Datasheet for 009-0034 Human Transferrin Rhodamine

## **Overview**

Description:	Human Transferrin Rhodamine Conjugated - 009-0034
Item No.:	009-0034
Size:	1 mg
Applications:	Dot Blot, IF, Multiplex
Origin:	Human

## **Product Details**

Background:	Human transferrin is encoded by the TF gene and is an iron-binding blood plasma glycoprotein that controls the level of free iron in biological fluids. Human transferrin binds iron very tightly but reversibly. Human transferrin is the most important iron pool in mammals. Human transferrin has a molecular weight of around 80 kDa and contains 2 specific high-affinity Fe(III) binding sites. The affinity of Human transferrin for Fe(III) is extremely high but decreases progressively with decreasing pH below neutrality.
Synonyms:	Human transferrin Rhodamine conjugation, TRITC conjugated transferrin
Species of Origin:	Human
Conjugate:	Rhodamine (TRITC)
Format:	Transferrin
Туре:	Native Protein
F/P Ratio:	2.27

## **Target Details**

Purity/Specificity:	This product was prepared from normal serum by delipidation, salt fractionation, selective precipitation and ion exchange chromatography followed by extensive dialysis against the buffer stated above. Assay by immunoelectrophoresis resulted in a single precipitin arc against anti-Human Transferrin and anti-Human Serum.
Relevant Links:	• GenelD - 7018
	UniProtKB - O06AH7



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#### • NCBI - AAB22049.1

## **Application Details**

<b>Tested Applications:</b>	Dot Blot
Suggested Applications:	IF, Multiplex (Based on references)
Application Note:	Human transferrin rhodamine conjugation has been tested by dot blot and is designed for immunofluorescence microscopy, fluorescence based plate assays (FLISA), fluorescent western blotting, multiplex analysis, including multicolor imaging, utilizing various commercial platforms.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.

## Formulation

Physical State:	Lyophilized
Concentration:	1.0 mg/mL by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	10 mg/ml Polyethylene Glycol (PEG-8000)
<b>Reconstitution Volume:</b>	1.0 mL
<b>Reconstitution Buffer:</b>	Restore with deionized water (or equivalent)

## **Shipping & Handling**

Shipping Condition:	Ambient
Storage Condition:	Store vial at 4° C prior to restoration. For extended storage aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.
Expiration:	Expiration date is one (1) year from date of receipt.

## Images



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#### Immunofluorescence Microscopy

COMMD1–PtdIns(4,5)P2 interaction measured by fluorescence quenching of all aromatics. (C) Example images showing colocalization of ATP7B with wild type, T174M and K167/173E COMMD1. ATP7B is in green and COMMD1 variants in magenta as indicated. Fig. 6.. PMID: 31515276.



#### Immunofluorescence Microscopy

Colocalization of ATP7B with TGN46 or LAMP1. Cells were left untreated (basal medium), treated with 10  $\mu$ M TTM (low copper) or treated with 10, 100 or 200  $\mu$ M CuCl2. (A,B) Merged images show ATP7B in green, TGN46 (A) or LAMP1 (B) in magenta and the nucleus in blue; pixel overlap is shown in white. Fig. 1. PMID: 31515276.



#### Immunofluorescence Microscopy

Rab5c over activation leads to excessive endolysosomal trafficking of Notch ligands and receptor.(A) TRITCconjugated TF internalization assay in Hela cells transfected with control empty pCS2 or pCS2-rab5c CA plasmid. Representative pictures were shown. Scale bar, 10  $\mu$ m. (B) Quantitative fluorescence intensity of intracellular TRITC-TF in control empty pCS2 and pCS2-rab5c CA transfected cells, n = 8 cells for each group. Error bars, mean  $\pm$  SD. P value was calculated by Student t test, \*\*\*P < 0.001. CA, constitutively active; TF, transferrin; TRITC, tetramethylrhodamine. S6 Fig. PMID: 32275659.

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#### Immunofluorescence Microscopy

Rab5c function is in an EC autonomous manner.(A) TRITCconjugated TF internalization assay in Hela cells transfected with empty pCS2 or pCS2-rab5c DN plasmids. Representative pictures were shown. Scale bar, 10  $\mu$ m. (B) Quantitative fluorescence intensity of intracellular TRITC-TF in empty pCS2 or pCS2-rab5c DN transfected Hela cells, n = 8 cells for each group. Error bars, mean ± SD. P value was calculated by Student t test, \*\*\*P < 0.001. DN, dominant-negative; TF, transferrin; TRITC, tetramethylrhodamine. S3 Fig. PMID: 32275659.

#### **Dot Blot**

Dot Blot of Rhodamine Conjugated Human Transferrin. Dotted directly with Rhodamine Conjugated Human Transferrin at following concentrations. Load: Lane 1 - 50ng Lane 2 - 16.67ng Lane 3 - 5.56ng Lane 4 - 1.85ng Lane 5 -0.62ng Primary antibody: none Secondary antibody: none Block: MB-070 for 1 HR at RT.



### References

- Heng J et al. Rab5c-mediated endocytic trafficking regulates hematopoietic stem and progenitor cell development via Notch and AKT signaling. *PLoS Biol.* (2020)
- Stewart DJ, Short KK, Maniaci BN, Burkhead JL. COMMD1 and PtdIns(4,5)P2 interaction maintain ATP7B copper transporter trafficking fidelity in HepG2 cells. *J Cell Sci.* (2019)

## Disclaimer



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