gibco

CTS™ Vitronectin (VTN-N) Recombinant Human Protein

In partnership with Cellular Dynamics International

Catalog Number CTS279S3

Pub. No. MAN0015673 Rev. 4.0



WARNING! Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. Safety Data Sheets (SDSs) are available from thermofisher.com/support.

Product description

Gibco™ CTS™ Vitronectin (VTN-N) Recombinant Human Protein provides a defined surface for feeder-free culture of human pluripotent stem cells in CTS[™] Essential 8[™] Medium.

Contents and storage

Contents	Amount	Storage	Shelf life ^[1]
CTS™ Vitronectin (VTN-N) Recombinant Human Protein	1 mL (0.9 mg/mL)	Store at -60°C to -80°C	36 months

^[1] Shelf life duration is determined from date of manufacture

Procedural guidelines

 Thaw CTS[™] Vitronectin at room temperature for 5-10 minutes, then place on ice.

Note: CTS[™] Vitronectin can be divided into usage-size aliquots in polypropylene tubes and stored at -60°C to -80°C.

- Room temperature storage and/or shaking might result in an appearance of light turbidity. This does not impact product performance.
- Cells cultured in CTS[™] Essential 8[™] Medium on CTS[™] Vitronectin-coated culture vessels should be passaged using CTS[™] Versene Solution, which is 0.48 mM EDTA in PBS.
- CTS[™] TrypLE[™] Select Enzyme along with CTS[™] RevitaCell[™] Supplement can be used for performing single-cell
- See Application Note on single-cell passaging methods from thermofisher.com/revitacell

Culture conditions

Culture type: Adherent feeder-free

Substrate: CTS[™] Vitronectin (VTN-N) Recombinant Human

Media: CTS[™] Essential 8[™] Medium Temperature range: 36°C to 38°C Incubator atmosphere range: Humidified atmosphere of 5% CO₂. Ensure that proper gas exchange is achieved in culture vessels.

Calculate the working concentration

The optimal working concentration of CTS[™] Vitronectin is cell line dependent and must be determined empirically. We recommend using an initial coating concentration of 0.9 µg/cm² for human pluripotent stem cell culture.

Prior to coating culture vessels, calculate the working concentration of CTS[™] Vitronectin using the formula below and dilute the stock appropriately. For the culture surface area and volume required, see "Culture surface area and volumes".

To coat a 6-well plate at a coating concentration of 0.9 µg/cm², you will need to prepare 6 mL of diluted CTS[™] Vitronectin solution (10 cm²/well surface area and 1 mL of diluted CTS[™] Vitronectin/well; see Table 1) at the following working

Working conc. = 0.9
$$\mu$$
g/cm² x $\frac{10 \text{ cm}^2}{1 \text{ mL}}$ = 9 μ g/mL

Dilution factor =
$$\frac{0.9 \text{ mg/mL}}{9 \mu\text{g/mL}}$$
 = 100X (i.e. 1:100 dilution)

Culture surface area and volumes

Table 1 Reagent volumes (in mL per well or per dish)

Culture vessel	Approximate surface area	Volume of diluted CTS™ Vitronectin solution
6-well plate	10 cm ² per well	1.0 mL per well
12-well plate	4 cm ² per well	0.4 mL per well
24-well plate	2 cm ² per well	0.2 mL per well
35-mm dish	10 cm ²	1.0 mL
60-mm dish	20 cm ²	2.0 mL
100-mm dish	60 cm ²	6.0 mL
T-25 flask	25 cm ²	2.5 mL
T-75 flask	75 cm ²	7.5 mL

Coat culture vessels with CTS[™] Vitronectin (VTN-N) Recombinant Human Protein

Instructions for coating a 6-well culture plate at a coating concentration of 0.9 $\mu g/cm^2$ are provided in this section. For volumes used in other culture vessels, see "Culture surface area and volumes". To calculate the working concentration of CTS Vitronectin used with other coating concentrations and to determine the appropriate dilution factor, see "Calculate the working concentration".

To coat the wells of a 6-well plate, add 60 µL of CTS[™] Vitronectin into a 15-mL conical tube containing 6 mL of sterile CTS[™] DPBS without calcium chloride, without magnesium chloride at room temperature.

Note: This results in a working concentration of 9 μ g/mL (i.e., a 1:100 dilution). Do not store diluted CTSTM Vitronectin solution.

2. Add 1 mL of the diluted CTS[™] Vitronectin solution to each well of a 6-well plate. When used to coat a 6-well plate (10 cm²/well) at 1 mL/well, the final concentration will be 0.9 μg/cm².

3. Incubate the coated plates at room temperature for 1 hour.

Note: The culture vessel can now be used or stored at 2°C to 8°C, wrapped in laboratory film, for up to one week. Do not allow the vessel to dry. Prior to use, pre-warm the culture vessel to room temperature.

 Aspirate the CTS[™] Vitronectin solution and discard immediately prior to use. Do not allow the culture surface to dry.

It is not necessary to rinse off the culture vessel after the removal of CTS[™] Vitronectin. Cells can be passaged directly onto the CTS[™] Vitronectin-coated culture vessels.

Related products

Unless otherwise indicated, all materials are available through thermofisher.com.

Item	Source
CTS™ Essential 8™ Medium	A2656101
CTS™ DPBS without calcium chloride, without magnesium chloride	A1285601
CTS™ Versene Solution	A4239101
CTS™ TrypLE™ Select Enzyme	A1285901
CTS™ RevitaCell™ Supplement	A4238401

Limited product warranty

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Revision history: Pub. No. MAN0015673 4.0

Revision	Date	Description	
4.0	27 November 2023	Updated Shelf life from 24 months to 36 months.	
3.0	8 February 2022	Updated Procedural guidelines and Calculate the working concentration topics with minor edits.	
2.0	6 July 2021	 Updated the product name. Updated to the current document template, with associated updates to the limited license information, trademarks, and logos. 	
1.0	29 September 2016	Baseline for this revision	

The information in this guide is subject to change without notice.

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